blocking compound. The sodium channel blocking compound can be one selected from the group of tetrodotoxin, anhydrotetrodotoxin, tetrodaminotoxin, methoxytetrodotoxin, ethoxytetrodotoxin, deoxytetrodotoxin and tetrodonic acid.

Pan '975 pertains to the use of amino hydrogenated quinazoline compounds for the treatment of drug dependence. Pan '975 fails to disclose or suggest in an enabling fashion the utilization of a sodium channel blocker such as tetrodotoxin for the treatment of pain. Further, Pan '975 fails to disclose the dosage, routes of administration, lack of adverse affects, and packaging as is set forth in the instant invention.

Pan '975 fails to suggest the present invention for at least two independent and distinct reasons set forth below, each one of which is sufficient of itself to demonstrate the patentability of the present invention.

I. Pan '975 is non-enabling for the treatment of pain

At page 2 of the Office Action, the Examiner states

> Pan et al. teaches the presently claimed compounds (column 3, line 36 - column 6, line 40) and that TTX, a prototypical example of such compounds, was known to be a potent analgesic for late cancer patients with no drug addiction occurring (column 2, lines 8-11) and that it was reportedly effective on relieving pain for lepers (column 2, lines 29-30).

However, Pan '975 at column 2, lines 7-11, relies upon the secondary source of Kao for the allegation that tetrodotoxin relieves pain:

> As a potent analgesic for late cancer patients, TTX exerted satisfactory effect of pain relieving on cancer pain, and no drug addiction cases were reported [Kao CY, Pharm. Rev. 18(2):997, 1966].

The Kao reference is discussed in the paragraph bridging pages 3 and 4 of the specification; a copy of the reference was filed with the IDS of February 16, 2001. As noted in the specification, Kao at page 1022 teaches away from the utilization of tetrodotoxin for the treatment of pain stating "specific actions of tetrodotoxin on central nervous system is debatable." See Kao at 1022, line 3. As a result, a person having ordinary skill in the art would be motivated by the allegations in Pan '975 to turn to the teachings of Kao, and subsequently discover that there is no rational basis to believe that tetrodotoxin can be utilized for the alleviation of pain.

Additional teachings against the utilization of tetrodotoxin to relieve pain (in the form of fugutoxin or fugufish extract) can be found in the S. Nomiyama reference that was referred to as reference number 117 in the Kao reference. A copy of the translation of the Nomiyama reference [NOMIYAMA S.: Pharmakologische Untersuchungen Über Fugugift, Folia Pharmacol. Jap. 95: 458-496, 1942 (Japanese)] is attached for the Examiner's convenience.

The Nomiyama reference at page 469, lines 1-2, asserts that fugutoxin has been used as a folk remedy on lepers (see Pan '975 at column 2, lines 29-30). However, the "tetrodotoxin" manufactured at the time of the reference (1942) was fundamentally different than the tetrodotoxin manufactured today.

The Sankyo "tetrodotoxin" manufactured during the 1940's was not a single compound, but a mixture of raw extract from fugu, or pufferfish, just like the fugutoxin mentioned in the Nomiya reference. This fugutoxin has a toxic strength and biological activity much lower than the tetrodotoxin known today (cf. e.g., claim 7).

Further, the Nomiyama reference states at page 496; "Tests of animals proved that tetrodotoxin does not possess remote analgesia effect, nor local analgesia effect. Neither has fugutoxin remote analgesia effect on human being." See Nomiyama at 496, lines 9-11. Thus, the Nomiyama reference, similar to Kao, teaches away from the presently claimed invention.

Yet further, Pan '975 at column 2, line 30 incorrectly cites the prior art. The reference referred to should be actually cited as Kao, CY, Pharmacology of Tetrodotoxin and Saxitoxin, Federation Proc. 31:1117-1123, 1972. However, this reference fails to teach anything about treating pain in lepers.

In order to better understand the non-enabling nature of Pan '975 for the treatment of pain, it is instructive to consider the

fundamentally different natures of cancer pain and the neuralgia associated with leprosy.

The nature of cancer pain is discussed as follows.

There are three types of pain based on where in the body the pain is felt: somatic, visceral and neuropathic. Pain of all three types can be either acute or chronic. Somatic, visceral and neuropathic pain can all be felt in the same individual at the same time, or they can be felt alone or at different times. Most cancer patients feel both somatic and visceral pain. Only about 15-20% of all cancer patients report neuropathic pain. The different types of pain respond differently to the various pain management therapies. Somatic and visceral pain are both much easier to manage than neuropathic pain.

Somatic Pain

Somatic pain is caused by the activation of pain receptors in either the cutaneous (the body surface) or deep tissues (musculoskeletal tissues). When it occurs in the musculoskeletal tissues, it is called deep somatic pain. Common causes of somatic cancer pain include metastasis in the bone (an example of deep somatic pain) and postsurgical pain from a surgical incision (an example of surface pain). Deep somatic pain is usually described as dull or aching but localized. Surface somatic pain is usually sharper and may have a burning or pricking quality.

Visceral Pain

"Viscera" is a word used to refer to the internal areas of the body that are enclosed within a cavity. Visceral pain is pain that is caused by activation of pain receptors from infiltration, compression, extension or stretching of the thoracic, abdominal or pelvic viscera (chest, stomach and pelvic areas). Common causes of visceral cancer pain include pancreatic cancer and metastases in the abdomen. This type of pain is not very well localized and is usually described as pressure-like, deep squeezing.

Neuropathic Pain

Neuropathic pain is caused by injury to the nervous system either as a result of a tumor compressing nerves or the spinal cord, or cancer actually infiltrating into the nerves or spinal cord. It also results from chemical damage to the nervous system that may be caused by cancer treatment (chemotherapy, radiation or surgery). This type of pain is severe and usually described as burning or tingling. Tumors that lie close to neural structures are believed to cause the most severe pain that cancer patients feel.

Acute Pain

Acute pain is short-lasting and usually manifests itself in very objective ways that can be easily described and observed. It may, for example, cause sweating or an increased heart rate. It can last for several days, increasing in intensity over time (subacute pain), or it can occur intermittently (episodic or intermittent pain).

Chronic Pain

Chronic pain is long-term and is defined as such if it lasts for over three months. It is much more subjective and not as easily described as acute pain. Effectively treating chronic pain poses a great challenge for physicians. This kind of pain usually affects a person's life in many ways. It can change somebody's personality, their ability to function, and their overall lifestyle.

Other Types of Pain

Pain is a very complicated physiological and psychological phenomenon that is not easy to categorize. There are several different ways to categorize pain. It can be somatic, visceral or neuropathic. It can be acute or chronic. Other terms used to describe pain include nociceptive, psychogenic, idiopathic, and referred. Nociceptive pain is pain that is sustained from ongoing tissue injury. It can be somatic or visceral, acute or chronic. Pain that is mainly psychological and not caused by actual tissue damage is called psychogenic pain. The fact that it is

> psychological does not make it any less painful or deserving of treatment than other types of pain. Idiopathic pain is pain that persists in the absence of any physical or psychological causes. Referred pain is pain that is felt in a part of the body that is far away from the actual site of what is causing the pain. See; www.oncologychannel.com/pain/types.shtml; See also www.asco.org/prof/pp/html/m_pain.htm (American Society of Clinical Oncologists).

The pain associated with cancer, discussed above, can be contrasted to the effects of Leprosy (Hansen's Disease).

> One of the main characteristics of Leprosy is its ability to affect the various nervous systems of the body, particularly the peripheral nerves. The key targets of M.leprae (Mycobacterium leprae) are the nerves' Schwann Cells. Leprosy does not affect the Central Nervous System. Where the sensory nerves are damaged, in varying degrees, they cannot register pain. Where those nerves supply the extremities of hands and feet, the latter are vulnerable to burns and other injuries that can often result in the loss of fingers and toes and sometimes hands and feet. Where the eye is affected, corneal anesthesia. Cranial Nerve involvement, can often lead to blindness, where the lack of health education makes the sufferer unaware of the means to prevent injury due to dust or other irritants. Where the motor nerves are involved, various forms of paralysis such as "Dropped Foot", "Dropped Wrist", "Clawed Hand", "Lagophthalmos" (eye cannot close due to nerve paralysis) can result. Where the autonomic nerves are damaged, the hair follicle, particularly in the cooler areas such as the eye-brows, can often result in the loss of hair in the affected parts. Damage to the autonomic nerves also can result in poor or no function of the sweat and sebaceous glands. This causes a drying of the skin and consequent cracking, exposing the sufferer to secondary infection. See www.web.raex.com/~bbeechy/introduction.html; See also Ramaratnam Sridharan et al., Neuropathy of Leprosy, eMedicine Journal, March 1, 2001, Vol. 2, No. 3, www.emedicine.com/neuro/topic266.htm.

Therefore, the neuralgic effects of leprosy are fundamentally different from the pain associated with cancer. Even if an analgesic substance is effective for leprosy pain, it will not necessarily be effective for cancer pain.

As discussed above, Pan '975 relies upon references that of themselves are non-enabling for the teachings for which the Examiner asserts the Pan reference. Furthermore, reading the references cited in Pan '975 would lead a person having ordinary skill in the art to believe that TTX is ineffective for the treatment of pain. Yet further, the Kao and Nomiyama references are non-enabling for dosage amounts, regimen and routes of administration for the treatment of pain.

"The evidentiary standard to be used throughout *ex parte* examination in setting forth a rejection is a preponderance of the totality of the evidence under consideration." In re Oetiker, 977 F.2d 1443, 1445, 24 USPQ2d 1443, 1444 (Fed. Cir. 1992). "After evidence or argument is submitted by the applicant in response, patentability is determined on the totality of the record by a preponderance of evidence with due consideration to persuasiveness of argument." In re Corkill, 771 F.2d 1496, 1500, 226 USPQ 1005, 1008 (Fed. Cir. 1985). A preponderance of the evidence exists when it suggests that it is more likely than not that the assertion in question is true. Herman v. Huddleston, 459 U.S. 375, 390 (1983).

Accordingly, the Examiner is respectfully requested to not only consider the teachings set forth in Pan '975, but to examine the totality of the evidence set forth in the teachings of Kao and Nomiyama.

In light of the above, it is clear that the teachings of Pan '975 are clearly erroneous. The treatment of erroneous data set forth in a reference is discussed in In re Yale, 434 F.2d 666, 168 USPQ 46 (1970) (listing in reference article of a chemical compound related to appellant's patent application was a typographical error).

> To the extent that anyone may draw an inference . . . that the mere printed conception or the mere printed contemplation which constitutes the designation of a 'compound' is sufficient to show that such a compound is old, regardless of whether the compound is involved in a 35 U.S.C. §102 or a 35 U.S.C. §103 rejection, we totally disagree. See 434 F.2d 666.

Thus, the mere statement in Pan '975 that treatment of pain from cancer or leprosy using TTX is old cannot lead to an inference such is truly the case.

As this rejection is based upon an erroneous and non-enabling teaching, withdrawal of the rejection is required.

II. Pan '975 fails to suggest the claimed embodiments

At page 2 of the Office Action, the Examiner admits that Pan '975 fails to disclose many of the embodiments of the present invention:

> The differences between the above and applicants' claimed subject matter lies in that the patentees do not highlight:
>
> (1) the presently claimed dosage amounts, regimens and routes of administration;
>
> (2) pain arising from other conditions than cancer and leprosy;
>
> (3) the presently claimed lack of adverse effects; and
>
> (4) the packaging as in present claim 22.

The Examiner then goes on to assert that these are "matters well within the purview of the skilled artisan." See Office Action at 3.

However, nowhere in the Office Action does the Examiner point out the teaching or suggestion in the Pan '975 reference itself for such embodiments as the dosage amounts, regimen, routes of administration, pain treatment conditions, lack of adverse effects and the packaging (cf. e.g., claims 3, 7, 8, 15-17, 19 and 22).

To establish a *prima facie* case of obviousness, it is necessary for the Examiner to present evidence, preferably in the form of some teaching, suggestion, incentive, or inference in the implied prior art, or in the form of generally available knowledge, that one having ordinary skill in the art would have been lead to use the relevant teachings of the implied references in the proposed manner asserted by the Examiner to arrive at the invention. See Ex parte Levengood, 28 USPQ2d 1300 (BPAI 1993). Because the Examiner bears the initial burden of presenting a

prima facie case of obviousness, if this burden is not met, then the burden of coming forth with evidence or argument does not shift to the Applicant. In re Rijckaert, 28 USPQ2d 1955 (Fed. Cir. 1993). Likewise, where an Examiner fails to establish a proper *prima facie* case, the rejection is improper, and should be overturned. In re Fine, 5 USPQ2d 1596 (Fed. Cir. 1988).

As has been shown, Pan '975 fails to be sufficient to produce *prima facie* obviousness for at least two independent and distinct grounds: 1) erroneous and non-enabling teachings; and 2) failure to present some teaching, suggestion, or incentive to produce the claimed embodiments of the present invention. Accordingly, Applicants believe withdrawal of this rejection under 35 U.S.C. §103(a) over Pan '975 is required.

Information Disclosure Statement

Applicants thank the Examiner for considering the Information Disclosure Statement filed February 16, 2001, and making the initialed form PTO-1449 of record in the application in the Office Action mailed May 4, 2001.

Conclusion

If the Examiner has any questions concerning this application, he is requested to contact Robert E. Goozner, Ph.D., Reg. No. 42,593, at (703) 205-8000 in the Washington, D.C. area.

If necessary, the Commissioner is hereby authorized in this, concurrent, and future replies, to charge payment or credit any overpayment to Deposit Account No. 02-2448 for any additional fees required under 37 C.F.R. § 1.16 or under 37 C.F.R. § 1.17; particularly, extension of time fees.

Respectfully submitted,

BIRCH, STEWART, KOLASCH & BIRCH, LLP

By _____ #32,568
Mark J. Nuell
Reg. No. 36,623

P. O. Box 747
Falls Church, VA 22040-0747
(703) 205-8000

DRN/REG/gml

[NOMIYAMA S.: Pharmakologischen Untersuchungen über Fugugift, Folia Pharmacol. Jap. 95:458-496, 1942 (Japanese)]

A Pharmacological Study of Fugutoxin

Shitsue Nomiyama

Advisers:
Prof. Fukuda, Director of Division of Pharmacology
Prof. Onotera, Director of Onotera Centre of Internal Medicine
Faculty of Medicine, Kyusyu Imperial University, Japan
Received on July 6, 1941

Table of Contents
Preface
Test Materials and Methods
Results
1. Relationship between General Reactions and Administration Routes
2. Analgesic effect
3. Effect on respiratory and circulatory systems
4. Effect on gastrointestinal tract (esp. vomitory effect)
Summary
Conclusions
References

Preface

Globefish is thought to be a special dish in Japan and China. The research in Fugutoxin, the toxin from globefish, has been carried out in Japan. Fugutoxin's pharmacological effect was studied long time ago by Osawa in the 18th year of Meiji, Dr. Takahashi and Dr. Inoko et al. in the 20th year of Meiji, respectively. Afterwards many reports about this study were issued by Hayashi, Takefuji, Karire, Itakura, Ishihara, Iwakawa, Kimura, Kunimasa, Yano, etc. It seems that the toxic action of Fugutoxin has been clearly recognized.

But the studies carried out before were primarily involved with Fugutoxin's effect on animals only. Therefore many unsolved questions still exist between the poisoning symptoms and course of human being by globefish and Fugutoxin's pharmacological effect on animals. For instance, among the two known major poisoning symptoms, perception paralysis and motor paralysis, it is not sure which will take place first or which is more severe; the cause of death is respiratory paralysis which, however, is not determined to be central paralysis or peripheral paralysis; many such questions remain still in dark. Nor has it been concluded that which pharmacological classification Fugutoxin falls into.

Recently the course and symptoms of intoxication of human being by globefish has been recognized through the relative study carried out by Fukuda and Tani, which emphasizes that the intoxication course goes very fast and vomiting is one of the symptoms that certainly happen. Such questions were not studied, like the intoxication course of animals by Fugutoxin, vomiting and the generative mechanism of vomiting, etc. This article will summarise my study of this subject under the advisement of Prof. Fukuda.

Material and Method of Experiment

Fugutoxin used in this experiment was extracted from the globefish's ovary by assistant Tani. To compare the effect, in some cases, Tetrodotoxin (manufactured by SANKYO) was also used.

Experimental animals included marmots, rabbits, cats, and dogs. To compare with the intoxication of human being, many cases were experimented on dogs and cats. Among the animals that have perception obstacle, the most sensitive ones, cats, were chosen to be experimentally wounded. Based on this experiment, clinical analgesia effect and the change of skin perception were studied.

Detailed experiment methods were discussed in each section.

Results

1. Relationship between General Reactions and Administration Routes

Many researches had indicated that the minimum lethal dose of Fugutoxin varies with the administration routes. According to Ishihara's study, for rabbits, the minimum lethal dose is 0.8ml (intravenous injection), 1.3ml(subcutaneous injection), 15.0ml (gastric ingestion). The lethal dose per kilogram does not apply to human body. Though not accurate, these administration routes was used to understand the basic of the Fugutoxin reaction. According to Iwakawa and Kimura, when Fugutoxin was given to rabbits by subcutaneous injection, rabbits would bend the body due to paralysis of respiration centre. When Fugutoxin was given by iv, paralysis of diaphragm nerve was observed. Reaction also varied with different density of Fugutoxin and the speed of absorbability. It was believed that the concentrate of Fugutoxin given by subcutaneous injection would cause paralysis of diaphragm nerve, and diluent of Fugutoxin given by iv would cause paralysis of centres. It is very important to understand the mechanism of reaction, the speed of absorbability and excretion of Fugutoxin under different administration routes.

Ishihara's experiment indicated that the intensity of the reaction of Fugutoxin varies with the species of animals. Based on the detailed experiment records of frogs, mice, and rabbits conducted by Iwakawa and Kimura, in addition to the intensity of reaction, qualitative changes were also observed. Therefore, I have experimented with marmots, cats, and dogs, administrating Fugutoxin per os, by iv, or subcutaneous injection. I wish I can clarify the mechanism of reaction of the Fugutoxin based on my observation of the general reaction and quantitative and qualitative differences. The experiments of this subject on these animals were extremely simple, the differences in administration routes was virtually not known.

a. Test in Marmots

Administration per os: Intubate catheter into marmot's stomach and inject Fugutoxin.

After the administration of Fugutoxin at 0.5ml/kg (volume of Fugutoxin to marmot body weight ratio) to the marmots, no toxic reactions were observed.

After being given 1.0ml Fugutoxin, the marmots indicated pink colour around their auricles, as well as dyscinesia in some cases.

10 minutes or so after being given 2.0 ml Fugutoxin, the marmots indicated red colour around their auricles, angeictasis around lips and then after 20 ~ 60 minutes indicated dyspneic respiration and dyscinesia. They squatted at corners with their abdomens supported by the ground. In most cases, the symptoms of marmots would not develop and be recovered in 2 ~ 3 hours. Notwithstanding, shortly minority marmots indicated slow and exaggerated respiration, and symptoms of dyspneic respiration as fanning their wings of nose and lower jaws. Also their limbs stretched and were not able to support their bodies. After being stimulated, they used forefeet to crawl with hind limbs dragged meanwhile. The above symptoms happened in 40 minutes after they were given Fugutoxin. If exacerbated further, their salivary secretion would increase and wet their lower jaws. Saliva stains could be found wherever their footprints were in the beds. Significant spanopnea happened to them as they were pumping air, and cyanosis took place around their lips and auricles. In 1.5 minutes, general convulsive seizure occurred and they used their lower jaws to bump the beds, or their heads faced upward, limbs stretched out with convulsive seizures. Some marmots died after 2 to 3 convulsive seizures.
After being given 2.5 ml Fugutoxin, the marmots had continuous angeictasis in their auricles, and indicated hypersalivation, motor paralysis, decrease in the tensility of skeletal muscles, pamplegia. They died in 1 to 1.5 hours after the administration. With their thoraces opened after their respiratory arrests, small amount of blood in black red could be observed. Their hearts continued beating for another 15 to 30 minutes till cardiac arrests occurred.
After being given 3.0 ml Fugutoxin with stronger action, they indicated respiratory arrests around 1 hour.
After being given 5.0 ml Fugutoxin, they indicated respiratory arrests more shortly.

Table 1. Administration of Fugutoxin per os (marmots)

Time	Sex	Body weight (kg)	Dose (ml/kg)	VSREAM*	Salivary secretion	DR***	Dyscine-sia	AAL**	Living or death#
27/II	F	0.51	0.3	-	-	-	-	-	L
27/II	F	0.51	0.3	-	-	-	-	-	L
28/II	F	0.60	0.5	-	-	-	-	-	L
28/II	F	0.69	0.5	-	-	-	-	-	L
1/III	M	0.56	1.0	-	##45 m.	30 m.	-	60 m.	L
1/III	F	0.60	1.0	-	-	45 m.	-	10 m.	L
I/III	F	0.59	1.0	-	-	-	-	-	L
2/III	F	0.50	2.0	-	10 m.	30 m.	-	25 m.	L
2/III	F	0.58	2.0	80 m.	40 m.	75 m.	60 m.	10 m.	115m L
2/III	F	0.59	2.5	-	45 m.	26 m.	42 m.	26 m.	L
5/III	M	0.60	2.5	80m.	43 m.	40 m.	40 m.	35 m.	90m L
5/III	F	0.75	2.5	71 m.	30 m.	28 m.	33 m.	28 m.	80m D
5/III	F	0.50	2.5	-	30 m.	20 m.	60 m.	17 m.	L
29/III	F	0.75	3.0	34 m.	27 m.	30 m.	40 m.	18 m.	56m D
29/III	M	0.50	3.0	45 m.	-	27 m.	42 m.	32 m.	50m D
29/III	F	0.65	3.0	50 m.	35 m.	35 m.	40 m.	-	70m D
1/V	M	0.75	5.0	27 m.	17 m.	20 m.	23 m.	25 m.	40m D
1/V	M	0.78	5.0	24 m.	19 m.	19 m.	24 m.	53 m.	27m D
1/V	M	0.74	5.0	30 m.	22 m.	20 m.	20 m.	25 m.	36m D

Note: * VSREAM: Absence of Skin Reflex of External Auditory Meatus
** AAL: Angeictasis in Auricles and Lips
*** DR: Dyspneic Respiration
# L: Living, D: Death
45 m. stands for the event occurred at the 45th min. after the administration of Fugutoxin. The similar as afterwards.

Subcutaneous injection: Subcutaneous injection at lateral abdomen.
After the administration of Fugutoxin at 0.05ml/kg (volume of Fugutoxin to marmot body weight ratio) to marmots, no toxic reactions were observed.

40 minutes after being given 0.1ml Fugutoxin, the marmots indicated angeictasis around their lips, algesthesias and auricles, as well as hypersalivation and dyscinesia. Later they indicated significant dyspneic respiration. In spite of these reactions, some of them recovered normal 3 hours later.
30 minutes or so after being given 0.2 ml Fugutoxin, the marmots indicated angeictasis and shortly, dyspneic respiration, dyscinesia, hypersalivation, cyanosis around lips and algesthesias, and platycoria. 1.5 to 2 hours after the administration, respiratory arrests occurred to them.
20 to 35 min. after being given 0.3 ml Fugutoxin, the marmots indicated the same symptoms as above. 1.5 hours later after the administration, respiratory arrests occurred to them.
About 70 min., 50 min., within 30 min. after being given 0.5 ml, 1.0 to 2.0 ml, 5.0 ml Fugutoxin, the marmots indicated respiratory arrests respectively. Their cardiac arrests came after their respiratory arrests did, the same with those given Fugutoxin via mouths. 10 to 20 min. before their respiratory arrests took place, their skin reflex of external auditory meatus vanished.

Table 2. Administration of Fugutoxin via subcutaneous injection (marmots)

Time	Sex	Body weight (kg)	Dose (ml/kg)	VSREAM*	Salivary secretion	DR***	Dyscine-sia	AAL**	Living or death#
4/III	M	0.62	0.05	-	-	-	-	-	L
4/III	M	0.58	0.05	-	-	-	-	-	L
6/III	M	0.65	0.1	-	-	-	120m	-	L
6/III	M	0.60	0.1	-	40m	52m	40m	-	L
6/III	M	0.60	0.1	-	-	78m	-	40m	L
8/III	F	0.71	0.2	-	42m	33m	40m	30m	L
8/III	F	0.73	0.2	75m	50m	60m	64m	47m	105m D
8/III	M	0.73	0.2	78m	73m	72m	92m	40m	92m D
13/III	M	0.62	0.3	80m	55m	60m	55m	20m	84m D
13/III	M	0.60	0.3	75m	63m	55m	70m	30m	93m D
13/III	M	0.60	0.3	82m	60m	121m	140m	-	90m D
13/III	M	0.61	0.3		68m	71m	60m	40m	L
13/III	M	0.63	0.3	-	70m	41m	85m	35m	85m D
13/V	F	0.79	0.5	50m	34m	45m	49m	22m	67m D
13/V	F	0.80	0.5	52m	39m	27m	43m	40m	62m D
13/V	F	0.83	0.5	40m	28m	35m	40m	32m	64m D
13/V	M	0.69	1.0	26m	47m	17m	26m	20m	57m D
15/V	F	0.70	1.0	24m	22m	17m	18m	14m	53m D
15/V	F	0.74	1.0	32m	20m	15m	25m	17m	55m D
25/V	F	0.50	2.0	22m	9m	7m	15m	10m	33m D
25/V	M	0.65	2.0	24m	10m	9m	17m	13m	40m D
25/V	M	0.75	2.0	18m	7m	7m	15m	10m	50m D
25/V	F	0.78	5.0	13m	7m	6m	13m	4m	23m D
25/V	F	0.74	5.0	14m	6m	9m	8m	7m	27m D

Note: * VSREAM: Absence of Skin Reflex of External Auditory Meatus
** AAL: Angeictasis in Auricles and Lips
*** DR: Dyspneic Respiration
# L: Living, D: Death
45 m. stands for the event occurred at the 45th min. after the administration of Fugutoxin. The similar as afterwards.

By administration per os or via subcutaneous injection, Fugutoxin may have general actions to various extents but may not have differences in action mechanism. Or to say, disturbance of perception, disturbance of respiration , circulatory disturbance and so will occur to both other marmots and human being in poisoning reactions to Fugutoxin. The symptoms indicated in premature stage are disturbance of respiration and peripheral angeictasis, which can be caused when about half the lethal dose is given to a subject. When the lethal dose is given to a subject, its disturbance of perception will not be observed until before its death. When approximate lethal dose is given to a subject per os, its dyscinesia will come shortly before its disturbance of perception did. When they are given Fugutoxin via subcutaneous injection minority marmots

indicate dyscinesia, and even if motor paralysis occurs to them, it will be slight. Several convulsive seizures will take place to a poisoning marmot before it dies. The convulsive seizure caused by administration of Fugutoxin per os is more remarkable than that caused by administration via subcutaneous injection. Their convulsion before death was caused by intravenous injection of Fugutoxin to rabbits in the test carried out by Iwakawa and Kimura. Followed by respiratory arrest, general convulsion can also be caused to mouse by subcutaneous injection of Fugutoxin, so can hypersalivation. As for poisoned marmots, no hemorrhage can be observed after their chest walls are opened. It is not the same at all with the bodies of other poisoned marmots. This may be resulted from collapse of blood vessels in chest wall caused by stasis of blood in some area due to paralysis of peripheral blood vessels.
The differences between the administration of Fugutoxin per os and that via subcutaneous injection: The ratio of their 50% lethal doses of subcutaneous injection is 0.2 : 2.5 = 1 : 12.5. The ratio of their minimum effective doses to cause disturbance of respiration and angeictasis is 0.1 : 1.0 = 1 : 10, approximately equal to the ratio of their lethal doses.

b. Test in Cats

Administration per os: Cat is a very sensitive animal. Without training, it is not easy to have a cat eat any food with Fugutoxin liquid but wooden fish that it likes. No other feed was provided on the day the test was performed.

No toxic reaction of cat was observed when 0.05 ml Fugutoxin per kilogram body weight was given.

After being given 0.1 ml/kg Fugutoxin per os, the cats indicated vomitive reactions and then inaction.

About 30min. after being given 0.2 ml/kg Fugutoxin per os, the cats indicated continuous 3 to 5 times vomiting, with food vomited at the beginning, and foam or only vomitive actions later. Sometimes they showed dread and uneasiness, and some of them displayed escape behaviour ceaselessly.

After being given 0.5 ml/kg Fugutoxin per os, the cats indicated significant symptom as vomiting. They vomited 5 to 6 times continuously at the beginning and intermittently later. 3 hours after the administration, they stopped vomitting. They were not so suffering while they were having vomit. If a cat could not vomit foam or bile, it would repeat vomitive action with its forefeet forcing so hard as to make its abdominal wall attach its back. Their claws seemed abnormal, while their forefeet and hind limbs tingled ceaselessly like they wanted to get rid of something from limbs. Notwithstanding, no changes happened to their algesthesia. Cyanosis were observed around their lips. In consecutive several days after, they suffered anorexia and some of them even died from hyposthenia.

About 30 min. after being given 1.0 to 3.0 ml/kg Fugutoxin per os, the cats displayed vomiting and hypersalivation. The duration of vomiting varied with the amount of Fugutoxin given per os, as generally from 1 hour to 2 hours. Some cats had their lips, algesthesias and auricles turn red one hour after the administration. 1.5 hours after the administration, they showed dyspneic respiration, uneasiness, excitation, hyperreflexia and some of them indicated tremor. A cat given per os a dose in this range displayed
dyscinesia, however, in 4 to 5 hours after they had stopped vomiting. The paralysis of forefoot came first and then that of hind limb as well as that of truncal muscles, finally general paralysis. Then it supinated, making neither any resistance nor any mewing sound, but it still had tendon

reflex. When cats were given 3.0 ml/kg Fugutoxin per os, such symptoms remained till the next day. They remained listless in the third to the 4th day ; On the 5th day they recovered a little and began eating. They displayed hypersalivation over a long time, with anorexia as well as gradually developed asthenia so that some of them died two weeks later. Under the above conditions of severe intoxication, the cats still indicated algesthesia even hyperalgia when examination was performed with their back skin being cut.

In oral administration of Fugutoxin to cats, the toxic reaction sequences were not parallel to the given amount per os. This may result from vomiting by which the cats threw out some or all toxin.

Table 3. Administration of Fugutoxin in cats per os

Time	No.	Sex	Body weight (kg)	Dose (ml/kg)	Analge-sia	Vomiting	Dyscine-sia	AAL**	Living or Death#
27/II	3	F	2.5	0.05	-	-	-	-	L
7/II	1	F	2.75	0.05	-	-	-	-	L
24/II	3	F	2.5	0.1	-	-	-	-	L
8/II	1	F	2.75	0.1	-	-	-	-	L
20/VI	4	F	3.3	0.1	-	50m	-	-	L
25/II	3	F	2.3	0.2	-	35-70m	-	-	L
24/VI	4	F	3.3	0.2	-	30-67m	-	-	L
10/II	1	F	2.7	0.2	-	27-200m	-	120m	L
2/III	3	F	2.0	0.5	-	30m	250m	130m	L 9/III D
26/VI	4	F	3.1	0.5	-	73-175m	-	-	L
14/II	1	F	2.4	0.5	-	62-100m	-	-	L
28/VI	4	F	3.2	1.0	-	30-105m	-	75m	L
18/II	1	F	2.2	1.0	-	65-120m	250m	65m	L
2/VII	4	F	2.8	1.5	-	30-42m	175m	45m	L
20/II	1	F	2.2	1.5	-	60-240m	300m	70m	L 4/III D. from asthenia
9/X	5	F	3.7	2.0	-	37-170m	260m	60m	L
19/XI	6	F	2.4	2.0	-	30-150m	-	40m	L
27/II	2	M	2.5	2.0	-	70-140m	270m	80m	L
14/VI	2	M	2.3	3.0	-	60-200m	310m-27h	120m	L
17/VI	2	M	2.1	3.0	-	65-175m	230m	100m	L
21/XI	6	F	2.5	3.0	-	45-220m	230m-25h	?	L 20/VI D

Note:

** AAL: Angeictasis in Auricles and Lips

L: Living, D: Death

45 m. stands for the event occurred at the 45th min. after the administration of Fugutoxin. The similar as afterwards.

Subcutaneous injection: No toxic reaction of cats was observed when they were given Fugutoxin 0.02 ml per kilogram body weight.

10 to 40 min. after being given Fugutoxin 0.02ml/kg by subcutaneous injection, the cats vomited 1 to 3 times.

After being given Fugutoxin 0.05 ~ 0.1 ml/kg by subcutaneous injection, the cats vomited nearly every time and continued 5 to 10 min., then changed into intermittent vomiting lasting 3 hours. Angeictasis in lips, auricles and so, and dyscinesia were indicated.
After being given Fugutoxin 0.25 ~ 0.3 ~ 0.5 ml/kg by subcutaneous injection, the cats vomited, and displayed uneasiness, tremor, hyperreflexia (increased quadriceps reflex) and angeictasis. At the beginning the cats indicated respiratory distress, then oligopnea and exaggerated respiration, dyscinesia along with absence of the tensility of skeletal muscles, and pamplegia. When a cat was hung with its back being hold, its trunk looked like a piece of pelage suspending. Notwithstanding, it did not indicate that the algesthesia had vanished, since some recovered as their motor paralysis disappeared on the next day. Though some cats recovered, their asthenia

exaggerated gradually so that they died after 10 days. Some other cats died about 1.5 ~ 4 hours after the administration.

After being given Fugutoxin 1.0 ml/kg by subcutaneous injection, the cats vomited in several minutes. After 20 min., dyspneic respiration and dyscinesia were indicated. Then oligopnea and cyanosis in lips, algesthesia and auricles were observed. Shortly platycoria, dyscinesia, general tremor, and analgesia appeared, and respiratory arrest occurred in about 30 minutes. The cardiac arrest took place in 20 to 55 minutes later.

Table 4. Administration of Fugutoxin in cats by subcutaneous injection

Time	No.	Sex	Body weight (kg)	Dose (ml/kg)	Absorption Analge-sia	Vomiting	Dyscine-sia (DY)	AALA **(A)	Living or Death#
9/II	1	M	2.8	0.01	-	-	-	-	L
3/X	4	F	2.7	0.01	-	-	-	-	L
23/X	4	F	2.8	0.01	-	-	-	-	L
11/II	1	M	2.7	0.02	-	-	-	-	L
5/X	4	F	2.7	0.02	-	10-22m	-	-	L
10/X	5	M	2.7	0.02	-	40m	-	-	L
15/II	1	M	2.5	0.05	-	5-210m	120m	-	L
4/III	2	F	3.2	0.05	-	5-200m	-	33m	L
15/V	3	F	2.5	0.05	-	30-55m	-	40m	L
17/II	1	M	2.5	0.10	-	10-30m	60-120m	27m	L
28/X	4	F	2.5	0.1	-	14-42m	58-140m	30m	L 3/XI D from Asthenia
20/V	3	F	2.4	0.1	-	11-30m	90m	-	L
9/III	2	M	3.2	0.2	-	10-170m	65-200m	60m	L
25/V	3	F	2.4	0.2	-	12-35m	130-270m	140m	L
11/X	4	F	2.6	0.2	-	9-20m	100-190m	47m	L
22/XI	1	M	2.3	0.25	113	10-95m	83m	80m	238m D
11/III	2	F	3.0	0.25	180	6-29m	58-190m	90m	L 2/III D from Asthenia
27/V	3	F	2.4	0.25	-	10-30m	120-240m	45m	L
12/III	6	F	2.0	0.3	105	10-40m	88-300m		L
13/III	7	F	1.8	0.3	98	7-32m	72m		120m D
14/III	6	F	2.0	0.3	120	10-75m	85DY110m	65m	182m D
13/V	9	M	2.0	0.5	78	10-75m	54DY80m	30m	95m D
15/V	3	F	2.2	0.5	45	4-17m	33m	25m	55m D
31/V	3	F	2.3	0.5	43	5-8m	37m	?	70m D
13/X	5	F	2.6	1.0	25	4-9m	23m	?	32m D
15/X	30	F	1.5	1.0	23	3-7m	20m	15m(A)	28m D

Note:

** AALA: Angeictasis in Auricles, Lips and Algesthesia.

L: Living, D: Death

45 m. stands for the event occurred at the 45th min. after the administration of Fugutoxin. The similar as afterwards.

General reactions: The most significant toxic reaction of cat to Fugutoxin is vomiting. Cats will vomit being given either per os or by subcutaneous injection, even at minim dose. They will also indicate hypersalivation before and after they show the vomitive symptoms. The second significant toxic reactions are dyscinesia, respiratory disturbance, and angeictasis. They display dyscinesia so extremely slowly that this symptom will come some time after vomiting does. Their dyscinesia lasts hours. The general motor paralysis may last as long as 20 hours after the administration per os. When their dyscinesia reaches its climax, they will indicate pamplegia with complete absence of tensile force. Therefore, they will not demonstrate spasm before death. Nonetheless their tendon reflex still doesn't vanish at all at the mean time. Vomiting happens no sooner than Angeictasis does. When larger dose is given by subcutaneous injection, angeictasis will still remain even after vomitive symptoms disappear.

Analgesia will not happen when Fugutoxin is given to cats per os. Only when Fugutoxin is given by subcutaneous injection at lethal or approximately lethal dose , it will occur before a subject's death. Nonetheless the pain reflex is even exaggerated at the beginning of intoxication when the injection dose is smaller than the lethal dose. Their limbs tingle in a manner like they want to get rid of something. When a cat is given Fugutoxin by subcutaneous injection, it may die as early as 30 to 60 min. after, or as late as 3 to 4 hours after. When lesser doses are injected, some cats will die in a week.

Being given Fugutoxin per os, those that died, which was not common, mostly would die in several days or a dozen or so days. The intoxication course of a human being evolves quickly, as his death will generally happen within 8 hours according to the report by Prof. Fukuda et al. He will mostly recover if he can survive this period. The intoxication course of a cat also evolves quickly, as it will die in 3 to 4 hours after being given Fugutoxin by subcutaneous injection. Notwithstanding, there are exceptions as some cats will die in a week after the administration.

The difference between per os and subcutaneous injection: It is very difficult to poison a cat by administering Fugutoxin per os. No cat died within several hour after the administration per os. This might lie in that they threw out most of the given toxic substance by vomiting so that no drastic intoxication could be caused to them. In only 2 to 3 cases, the subjects developed asthenia by abstinence of food, and died in several days or a dozen or so days.

Of administration by subcutaneous injection, the 50% lethal dose and the vomitive dose are 0.3 ml/kg. The least vomitive dose: that of subcutaneous injection is 0.02 ml/kg, and that of per os is 0.2 ml/kg.

Vomiting time: 5 min. as large dose is given by subcutaneous injection, and 10 min. as small dose is. As for administration per os, most subjects vomited within 30 to 60 min after the administration. Based on these indications, the reaction is not caused by gastric reflex but cerebral vomiting.

c. Test in Dogs

Administration per os: The toxicant was mixed with 300 gram of feed so as to have dogs eat the mixture willingly.

No toxic reactions were observed after 0.3 ml/kg Fugutoxin was given to the dogs.

Vomitive reactions were observed in 80 min. or so after 0.5 ml/kg Fugutoxin was given.

Vomitive reactions would certainly be observed in about 60 min. after 0.7 ml/kg Fugutoxin was given. A subject vomited four times in the first 20 min., and then vomited only foam.

After 1.0 ml/kg Fugutoxin was given, the subjects vomited in 40 min. to 3 hours, continuously at the beginning and subsequently only with vomitive behaviour. Dyscinesia happened at the 2nd hours after the administration with inaction. Some subjects could recover if their respiratory disturbance was slight.

After 30 to 60 minutes 2.0 ml/kg of Fugutoxin was given, the subjects started vomitting, at first slightly, then continuous vomiting, which is unique symptom of Fugutoxin intoxication, and then turned into intermittent vomiting. They stopped vomiting 1.5 to 2 hours after the administration. Hypersalivation, hyperreflexia and dyspneic respiration occurred. About one hour after, they tried to maintain erect position by leaning against walls due to motor paralysis. Shortly they fell down when dyscinesia took place, with platycoria and oligopnea. The duration of inspiration was longer, as three times that of expiration. Some of them had respiratory arrests subsequently.

After 3.0 ml/kg Fugutoxin was given, the subjects indicated frequent vomiting in a period of 20 to 30 min. About 20 min. later motor paralysis occurred, and so did very shortly respiratory inhibition with platycoria. They died in about 50 min. With their chests opened, observed was small amount of blood in dark red without bleeding, with their hearts still beating. Hear arrests took place 10 to 20 min. later.

Administration by endogastric administration: When the stomach of a subject was empty, the toxic substance was forced into it through a stomach tube.

5 to 30 min. after 1.0 ml/kg Fugutoxin was given to dogs, they indicated frequent vomiting with uneasiness, and being in excitatory state. Shortly observed were dyspneic respiration, dyscinesia, general tremor, analgesia, and death.

Table 5. Administration of Fugutoxin in dogs per os

Time	No.	Sex	Body weight (kg)	Dose (ml/kg)	Absorption Analge-sia	Vomiting	Dyscine-sia (DY)	AAL **(A)	Living or Death#
16/III	1	F	11.0	0.2	-	-	-	-	L
16/III	1	F	11.0	0.2	-	-	-	-	L
21/III	2	M	8.0	0.3	-	-	-	-	L
16/III	1	F	11.0	0.3	-	-	-	-	L
16/III	1	F	10.9	0.3	-	-	-	-	L
3/X	3	M	26.0	0.5	-	-	-	-	L
4/X	3	M	27.0	0.5	-	80m*	-	-	L
21/III	2	M	8.0	0.5	-	-	-	-	L
5/X	3	M	27.0	0.7	-	60-95m	-	-	L
23/III	1	F	10.5	0.7	-	65-85m	-	90m	L
8/X	2	M	8.5	0.7	-	75-100m	-		L
7/X	3	M	26.2	1.0	-	44-126m	70m	?	L
10/X	3	M	26.2	1.0	-	55-107m	65m		L
9/X	2	M	8.0	1.0	-	158-225m	-		L
13/X	2	M	8.0	2.0	-	68-98m	49m		L
12/X	3	M	26.0	2.0	-	37-125m	60m		L
15/IV	1	M	10.5	2.0	-	32-66m	54m	?	78m D
14/IV	4	F	11.0	3.0	53m	24-45m	48m		65m D
13/IV	2	F	8.2	3.0	40	28-40m	41m		43m D
24/III	5	M	7.1	1.0	35m	5-24m	30m		42m D
14/X	6	F	9.7	1.5	-	6-26m	78m		L
16/X	7	F	7.0	2.0	32m	5-20m	27m		40m D
15/X	6	F	9.0	3.0	30m	5-28m	24m		38m D

Note:

* Vomitive behaviour

? Not clear in the original report by Japanese.

** AAL: Angeictasis in Auricles, Lips.

L: Living, D: Death

45 m. stands for the event occurred at the 45th min. after the administration of Fugutoxin. The similar as afterwards.

After 1.5 ml/kg Fugutoxin was given, the dogs indicated frequent vomiting, and dyscinesia in 80 min. without analgesia, and then recovered.

After 2.0 ~ 3.0 ml/kg Fugutoxin was given, the dogs indicated continuous vomiting, and vomited only foam in 20 min. Also observed were uneasiness, dyspneic respiration, dyscinesia as well as shaking erect position. They fell down in 30 min, shortly demonstrated platycoria, analgesia, and respiratory arrests in about 40 min.

Administration by subcutaneous injection: No toxic reactions were observed after Fugutoxin was given at 0.005 ml per kilogram body weight.

4 to 27 min. after 0.01 ml/kg Fugutoxin was given, dogs indicated vomiting, slight uneasiness. But they usually recovered after 1 hour.

After being given Fugutoxin 0.02 ml/kg by subcutaneous injection, in several minutes the subjects indicated vomiting, uneasiness, hyperreflexia, platycoria, hypersalivation, sometimes general paralysis, and respiratory disturbance. But they recovered 2 to 5 hours later.

After being given Fugutoxin 0.03 ~ 0.05 ml/kg by subcutaneous injection, the subjects indicated frequent vomiting in a period of several minutes to 20 and more minutes. Later the vomiting changed to be intermittent, and halted in 60 ~ 90 min. In 80 min. or so salivary secretion increased along with general tremor, platycoria, and hyperreflexia. Shortly they displayed staggering gait with hind limbs dragged, and then paralysis of forefeet and head ptosis in abdominal horizontal position. They also indicated slight respiratory inhibition. 3 hours later, some recovered a little but some other had respiratory arrests, felling down like tumbling with respiratory arrests shortly.

Table 6. Administration of Fugutoxin in dogs by subcutaneous injection

Time	No.	Sex	Body weight (kg)	Dose (ml/kg)	Absorption Analge-sia	Vomiting	Dyscine-sia (DY)	Tremor	Living or Death#
20/III	1	M	8.5	0.003	-	-	-	-	L
20/III	1	M	8.5	0.003	-	-	-	-	L
22/III	1	M	8.6	0.005	-	-	-	-	L
22/III	1	M	8.6	0.005	-	-	-	-	L
11/IX	2	M	17.8	0.005	-	-	-	-	L
14/X	3	F	7.4	0.005	-	4m	-	-	L
16/X	4	M	9.0	0.005	-	-	-	-	L
25/VI	5	F	8.6	0.01	-	4-66m	-	-	L
11/IX	3	M	7.3	0.01	-	29m	-	-	L
25/III	5	F	8.6	0.01	-	-	-	-	L
12/IX	3	F	7.2	0.02	-	10-60m	-	-	L
13/IX	2	M	17.9	0.02	-	15-65m	-	-	L
27/III	3	F	7.0	0.02	-	8-40m	40-170m	45m	L
14/IX	6	F	7.0	0.03	-	5-45m	70-310m	90m	L
29/III	7	F	6.8	0.03	78m	10-50m	52m	80m	180m D
15/IX	6	F	7.0	0.03	-	5-50m	60-250m	78m	L
16/IX	5	M	8.4	0.05	-	4-105m	50-280m	95m	L
17/IX	5	M	8.4	0.05	-	5-25m	55-180m	75m	L
18/IX	5	M	8.2	0.05	130m	6-78m	52-195m	78m	150m D
25/IX	2	M	15.1	0.06	-68m	10-42m	42-85m	5m	78m D
26/IX	1	M	8.9	0.06	62m	3-18m	32m	58m	75m D
27/IX	6	F	7.0	0.06	76m	4-20m	38m	70m	92m D
21/VI	6	F	6.3	0.1	27m	2-14m	14-27m	14m	27m D

Note:

L: Living, D: Death m: minute

After being given Fugutoxin 0.06 ml/kg, the dogs indicated frequent drastic vomiting in a period of several minutes to 30 min., and uneasiness, along with excitatory state. In 40 min. displayed were dyspneic respiration, dyscinesia, and afterward general tremor, platycoria, analgesia. Deaths took place in about 80 min.

After being given Fugutoxin 0.1 ml/kg, the dogs indicated instantly vomiting for several minutes, and dyspneic respiration, uneasiness, excitatory state, hyperreflexia. Shortly motor paralysis occurred with only their heads labile in about 20 min. Deaths took place in 27 min.

General reactions: When a dog is poisoned, among the first symptoms are vomiting and increase of salivary secretion, similar with a cat being poisoned. Notwithstanding, a dog will indicate

uneasiness, excitatory state and tremor, hyperreflexia, and platycoria prior to motor paralysis, which will slowly take place following these excitatory states and vomitive symptoms. Even at last motor paralysis was so insignificant that some dogs could still remain erect position.

Dog's perception is so optuse that it is difficult to conduct perception examination to a dog. In the telophase of severe intoxication caused by injection of approximate lethal dose of Fugutoxin to it, a dog had no reaction to strong stimulation by compression. This meant unconsciousness in the telophase of intoxication.

The difference between administration of the mixture of toxin liquid and feed through stomach tube to empty stomach and that per os: Of the former, the 50% lethal dose is 3.0 ml/kg; of the later, 1.0 ml/kg given can be lethal to the subject. As given per os, a dog might indicate vomiting in as early as 30 min; while given through stomach tube, it might do so in 5 min. As given through stomach tube, a subject displayed dyscinesia very soon. The reason was that the subject absorbed faster Fugutoxin given through stomach tube while its stomach was empty. It is the same with all drugs but more significantly with Fugutoxin.

The intoxication course of a dog evolves rapidly. Given Fugutoxin by subcutaneous injection or per os, generally a dog will die in 30 to 90 min, but some will decease in 3 hours. Cats don't undergo such persisting-poisoning death.

The difference between administration per os and that by subcutaneous injection: 50% lethal dose: for administration per os is 0.06 ml/kg and for that by subcutaneous injection, 3.0 ml/kg. The ratio of these two is 1:50.
Least vomitive dose: for administration per os is 0.01 ml/kg and for that by subcutaneous injection, 0.7 ml/kg. The ratio of these two is 1:70.
The ratio of marmot's lethal doses per os and by subcutaneous injection and that of cat's vomitive doses per os and by subcutaneous injection are both about 1: 10. While the ratio of dog's respective doses is even greater, possibly due to strong function of detoxification and weak absorption capacity of its alimentary canal.
Dog's 50% lethal dose by intra-venous injection is 0.06 ml/kg. It is noticeable that dog's lethal dose by intra-venous injection is nearly equal to that by subcutaneous injection. However, it takes a dog 20 min. to decease by iv. but 80 min. by subcutaneous injection. Based on the experimental results provided by Iwakawa and Kimura, the least lethal dose by iv for rabbit is 0.00075 ml/kg and that by subcutaneous injection is 0.0015 ml/kg, namely the difference between these two is the later twice the former.
That the lethal dose by iv is close to that by subcutaneous injection indicates that subcutaneous tissue can absorb toxic liquid smoothly.

2. Analgesic effect

In folklore Fugutoxin was alleged to possess analgesia effect; Tetrodotoxin has been used for decades particularly on the neuralgia suffered by lepers. Itakura, Iwakawa, Kimura and Yano et al carried out tests with reflex frogs and rabbits, which proved that Fugutoxin can produce anesthesia on cornea similar with that by cocaine. It has not been recognized whether Fugutoxin has remote effect or not except local effect. Itakura found out in his tests that Fugutoxin could cause perception paralysis in hind limbs of rabbits, as well as paralysis of motor nerve and absence of genual tendon reflex in them. Therefore, Fugutoxin can't give rise to absence of perception. Consequently, it has no pharmacological basis to administer Fugutoxin for clinical use as analgesic with remote effect. Nevertheless, up to now most tests has been performed with rabbits as subjects. According to the latest experimental results provided by Kasuka, marmot had so sensitive algesthesia that its skin reflex in external auditory meatus could be utilized to examine the local analgesia effect. Cat is also one of the very sensitive and acute animals which we can employ as subjects for the tests of Fugutoxin's analgesia effect.

The Fugutoxin liquid was prepared mainly by Tani, the assistant. Sinomenin, Veratrin and morphine are also used in control tests for the purpose of comparison. Clinical test was also performed in order to recognise whether or not Fugutoxin has analgesia effect on human beings.

a. Test in Marmots

As specified in Kasuka's test method, a piece of hair was employed as stimulant to irritate the skin of external auditory meatus, and the absence of auricle reflex was made a marker for examining Fugutoxin's analgesia effect.

Administration per os: as displayed in Table 1, neither reaction nor change in algesthesia was observed when Fugutoxin 0.5 ml per kilogram body weight was given. After Fugutoxin 1.0 ml/kg was given, observed were angeictasis in auricles, around mouth and at algesthesia, dyspneic respiration, motor inhibition, then hypersalivation, shortly respiratory inhibition. Notwithstanding, the skin reflex of external auditory meatus indicated no abnormal change. After Fugutoxin 2.0 ~ 2.5 ~ 3.0 ~ 5.0 ml/kg were given, observed were absence of muscular tensile force, motor paralysis, indicating symptoms of severe intoxication. Absence of skin reflex of external auditory meatus was also displayed when most subject animals died.

Administration by subcutaneous injection: As shown by Table 2, no reaction was found after Fugutoxin 0.05 ml/kg was injected. After Fugutoxin 0.1 ml/kg was injected, observed were angeictasis in auricles, around lips and at algesthesia, and then motor paralysis, but not abnormal change in skin reflex of external auditory meatus. After Fugutoxin 0.2 ml/kg was injected, drastic reactions were caused. Shortly after the absence of skin reflex of external auditory meatus the animals died. If it was not developed to the degree of reflex absence, an animal could recover to normal. After Fugutoxin 0.3 ~ 0.5 ml/kg was given, more drastic reactions were demonstrated and the animals deceased with reflex absence occurring before death.

Sinomenin: No reaction was observed when Sinomenin 0.05 ml per kilogram body weight was given by subcutaneous injection.

After Sinomenin 0.1 ~ 0.5 ~ 1.0 ~ 2.0 ~ 5.0 ml/kg was given, analgesia took place only in the injected local area. The animals showed no pain response when cut by a knife or pierced by a needle, but slight degree of dyscinesia. No absence of skin reflex of external auditory meatus was observed.

Veratrin: As shown in Table 7, no reaction was shown after Veration 0.2 mg per kilogram body weight was given by subcutaneous injection.

After Veratrin 0.3 mg/kg was given by subcutaneous injection, muscular spasm and contraction took place in about 90 min.

After Veratrin 0.5 mg/kg was given by subcutaneous injection, hypersalivation occurred in 12 to 55 minutes and the animals became inactive.

After Veratrin 1.0 mg/kg was given by subcutaneous injection, observed were hypersalivation, spasm and contraction, screaming, odontoprisis, dyspneic respiration, and dyscinesia in about 10 minutes, paralysis state at 20 min. with presence of skin reflex of external auditory meatus. Since 3 hours later, the animals had been slowly recovering to normal till the next morning.

After Veratrin 2.0 mg/kg was given by subcutaneous injection, drastic reactions happened rapidly. The animals screamed like their heads were tightened in more than 10 minutes, and indicated dyscinesia in about 12 min. with hypersalivation. They indicated uneasiness in 20 min. due to dyspneic respiration, turning around frequently, screaming when absence of skin reflex of external auditory meatus occurred. They deceased in an hour.

Table 7. Administration of Fugutoxin in marmots by subcutaneous injection

Time	Sex	Body weight (kg)	Dose ml/kg	Absorp-tion Analge-sia	Sali-vary secretion	Dyspeic respira-tion	Hyper-reflixi a	SMI*	Scream -ing	Muscu-lar spasm	Living or Death#
28/XI	F	0.55	0.2	-	-	-	-	-	-	-	L
28/XI	F	0.5	0.2	-	-	-	-	-	-	-	L
28/I	F	0.55	0.3	33m	-	-	-	-	-	90m	L
28/I	M	0.6	0.3	-	-	-	-	-	-	75m	L
29/I	F	0.6	0.3	-	-	-	-	-	-	80m	L
27/I	F	0.47	0.5	-	12-55m	-	--	12-45m	45m	-	L
27/I	M	0.5	0.5	-	15-50m	-		20-65m	-	-	L
26/I	M	0.52	0.5	-	20-65m	-	-	31-93m	4-5m	65m	L
26/I	F	0.47	1.0	-	8-90m	-	-	-	-	-	L
25/I	F	0.8	1.0	28m	10-90m	10m	-	31-81m	18-120m	46m	L
25/I	M	0.49	1.0	37m	15-80m	17m	-	22m	24m	52m	L
25/I	F	0.55	2.0	45m	10-20m	20m	27m	12m	20m	39m	63m D
24/I	F	0.5	2.0	45m	10-37m	20m	35m	15m	18m	45m	56m D
24/I	F	0.55	2.0	-	10m	12m	30m	17m	23m	30m	50m D
26/I	F	0.65	3.0	30m	12-15m	11m	-	12m	22m	22m	34m D
28/I	F	0.55	3.0	20m	3m	15m		14m	25m	24m	33m D

Note: * SMI: Spontaneous motor inhibition
L: Living, D: Death m: minute

When Fugutoxin was given to marmot per os or by subcutaneous injection, the absence of skin reflex of external auditory meatus would happen only before death. Therefore, Fugutoxin didn't have any special analgesia effect.

Sinomenin did not cause the absence of skin reflex of external auditory meatus but analgesia in the injected local area as no pain response indicated to cutting or pierceing with a needle.

Veratrin was able to cause absence of skin reflex of external auditory meatus but only when approximate lethal dose was given. Therefore, Veratrin had no analgesia effect.

As mentioned above, marmot was not eligible for examination of after-absorption analgesia. Although analgesia was not observed after Fugutoxin was given to marmots, it could explain that Fugutoxin did not have absorptive analgesia effect.

b. Test in Cats

The hair on the back of a well-trained cat was removed, and a incision was made in each side of its spine, then a sharp needle was employed to pierce and press the edge of an incision opening in order to examine the presence of pain response, based on its expression, crying, skin reflex of back, escape behaviour, fear, anger and the feeling of operator's hands and that on the operation of the needle.

In order to find out the presence of absorption function, the test liquid was injected into subcutaneous tissue in a area in the back but in a distance from the incisions. In order to examine the pain paralysis in local area, one side of the back was injected at first and then incision was made in the injected area, or a needle was employed to pierce and press around the opening of injection.

The incision of one side was painted pantocain as comparison. After being painted pantocain, the incision would not indicate any reactions mentioned above so that it was very easy to determine.

Fugutoxin

Administration per os: Totally 21 cases were displayed in Table 3.

After Fugutoxin was given at 0.5 ml per kilogram body weight, frequent vomiting was caused, as well as abnormal indications of limbs as they tingled forcefully like getting rid of something.

After Fugutoxin 1.0 ~ 1.5ml/kg was given, caused were frequent vomiting, hypersalivation, dyspneic respiration but absorptive analgesia or local algesthesia paralysis.

After Fugutoxin 2.0 ~ 3.0 ml/kg was given, caused were vomiting, uneasiness, hyperreflexia, tremor but analgesia or local algesthesia paralysis. Notwithstanding manifest motor paralysis, algesthesia became more acute rather than analgesia.

Administration by subcutaneous injection: Totally 26 cases in Table 4. After Fugutoxin was given at 0.1 ~ 0.2 ml per kilogram body weight, caused were frequent vomiting and then dyscinesia but remote or local analgesia.

After Fugutoxin 0.25 ~ 0.5 ml/kg was given, observed were platycoria, dyspneic respiration, and then decrease in muscular tensile force, significant dyscinesia, remote analgesia, and shortly respiratory arrest.

Tetrodotoxin

Administration by subcutaneous injection:
As shown in Case #31 in Table 8, no reaction was shown when less than 0.04 ml/kg Fugutoxin was given. When the dose was increased to 0.05 ml/kg, vomiting was caused in 10 ~ 30 minutes; in some cases, no reaction was observed.

At 0.15 ml/kg, vomiting started after around 8 minutes, then hypersalivation and angeictasis around their algesthesias and auricles after 35 minutes, but no remote or local analgesia.
At 0.2 ml/kg, except vomiting, no dyscinesia or dyspneic respiration was observed.
After 5 minutes of 0.23 ~ 0.3 ml/kg Fugutoxin was given, observed vomiting. After 20~60 minutes, hypersalivation, angeictasis, dyspneic respiration, dyscinesia, hyperreflexia, body thrilling were observed. After around 60 minutes, found cyanosis around lips and auricles; after 40~80 minutes, remote analgesia started. Some recovered algesia after 2~3.5 hours, some died after 2 hours, and some died after 3 days due to anorexia and asthenia.
At 0.5 ml/kg, frequent vomiting, dyscinesia, dyspneic respiration, corectasis, cyanosis around lips and auricles were observed after 4~15 minutes, then remote analgesia appeared, and respiratory arrest occurred in about 30 minutes. The cardiac arrest took place in 15 to 30 minutes later.

Table 8. Administration of Tetrodotoxin in cats by subcutaneous injection

Time	No.	Sex	Weight (kg)	Dose (ml/kg)	Absorptive Analgesia	Vomiting	Dyspneic respiration	Dyscinesia (DY)	AALA **(A)	Hyper-salivation	Living or Death#
29/V	1	M	3.5	0.01	-	-	-	-	-	-	L
28/X	3	M	9.8	0.01	-	-	-	-	-	-	L
9/II	2	M	2.8	0.01	-	-	-	-	-	-	L
29/V	3	F	2.5	0.02	-	-	-	-	-	-	L
11/II	2	M	2.7	0.02	-	-	-	-	-	-	L
30/V	1	M	3.2	0.03	-	-	-	-	-	-	L
1/VI	2	M	2.6	0.05	-	-	-	-	-	-	L
31/V	1	M	3.2	0.05	-	35m	-	-	-	-	L
30/XI	6	F	1.3	0.05	-	13m	23m	-	-	-	L
1/XII	6	F	1.3	0.1	-	8M	-	-	-	-	L
7/XII	5	F	2.2	0.1	-	6m	-	-	-	-	L
6/XII	7	F	2.3	0.1	-	8m	60m	-	-	-	L
2/XII	4	M	4.6	0.15	-	8m	-	-	35m	-	L
23/XII	7	F	2.1	0.15	-	5m	37m	-	48m	-	L
13/XII	7	F	2.0	0.17	48m	6m	20m	40m	-	-	L
16/XII	6	F	2.3	0.17	180m	11m	180m	90m	42m	-	L
14/XII	7	F	2.1	0.17	-	4m	72m	60m	-	-	L
7/XII	5	F	2.2	0.2	-	7m	-	-	45m	-	L
20/XII	5	F	2.2	0.2	-	9m	-	42m	-	-	L
2/II	1	M	3.2	0.2	-	5m	80m	120m	-	-	L
22/XII	6	F	2.2	0.23	53m	5m	33m	38m	Cyanosis 45m	20m	120m D
24/II	1	M	3.2	0.23	55m	5m	13m	30m	11m Cyanosis 45m	60m	L
13/II	3	F	2.5	0.23	-	10m	36m	-	-	27m	L
17/II	3	F	2.5	0.25	-	8m	43m	78m	-	-	L
10/II	4	M	4.5	0.25	80~120m	8m	50m	60m	60m	23m	L
23/II	6	F	2.5	0.25	-	6m	35m	55m	34m	19m	D (asthenia)
14/II	4	M	4.5	0.3	50~180m	7m	30m	40m	45m Cyanosis 55m	25m	L
20/II	4	M	4.3	0.3	40~200m	5m	30m	30m	37m Cyanosis 43m	?	L
19/XII	6	F	1.3	0.5	20m	4m	23m	13m	-	7m	36m D
29/XII	9	F	2.0	0.5	27m	4m	20m	17m	-	11m	42m D
30/XII	10	F	2.2	0.5	23m	3m	21m	16m	17m Cyanosis 21m	?	33m D

Note:

** AAL: Angeictasis in Auricles and Lips
# L: Living, D: Death

Table 9. Administration of Sinomenin hydrochloride to cat by subcutaneous injection

Time	No.	Sex	Weight (kg)	Dose (ml/kg)	Absorp-tive analgesia	Local analgesia	Hyper-salivation	Vomiting	Dyspneic respiration	Corectasis	Hyper-reflexia	Dyscine-sia	AALA **(A)	Defeca-tion	Urina-tion
28/IX	1	M	3.7	0.1	-	7~90m	-	-	-	-	48~140m	-	48~160m	1	-
28/IX	2	F	2.4	0.1	-	10~100m	-	-	-	-	35~130m	-	53~152m	1	-
29/IX	1	M	3.6	0.2	-	9~97m	-	-	-	-	45~145m	-	45~157m	-	-
1/X	2	F	2.4	0.2	-	10~80m	-	-	-	50~170m	15~160m	-	15~150m	1	-
1/X	3	F	2.6	0.2	-	10~90m	-	-	-	20m	20~130m	-	20~130m	2	-
2/X	2	F	2.3	0.5	-	10~180m paralysis 25m	10~63m	10m twice	20~170m	13~420m	22~125m	-	10~180m	1	2
3/X	1	M	3.4	0.5	-	15~150m paralysis 30m	12~90m	95m once	25~140m	5~150m	12~240m	-	10~240m	2	2
4/X	3	F	2.6	0.5	-	10~120m paralysis 25m	30~54m	25m twice	27~155m	15~180m	15~160m	-	20~137m	1	1
5/X	1	M	3.4	1.0	110m~?	10~150m	27~73m	30~127m 3 times	20~137m	20~170m	17~400m	37~95m	28~147m	2	1
6/X	2	F	2.3	1.0	-	12~173m	20~75m	25~75m twice	18~180m	21~185m	12~270m	42~110m	17~123m	1	2

Note: ** AAL: Angeictasis in Auricles and Lips

Table 10. Administration of Veratrin to cat by subcutaneous injection

Time	No.	Sex	Weight (kg)	Dose (ml/kg)	Absorp-tive analgesia	Hyper-salivation	Vomiting	Dyspneic respiration	Corec-tasis	Hyper-reflexia	Alysmus	Dyscine-sia	Screa-ming	Spasm	Diarrhea	Living or Death#
29/XI	1	M	1.8	0.02	-	-	-	-	-	-	-	-	-	-	-	L
1/XI	1	M	1.8	0.02	-	-	-	-	-	-	-	-	-	-	-	L
2/XII	2	F	2.2	0.02	-	-	-	-	-	-	-	-	-	-	-	L
2/XII	3	F	3.2	0.05	-	-	20m	-	-	23m	-	-	-	-	-	L
3/XII	4	F	3.7	0.05	-	-	25m	-	-	26m	-	-	-	-	-	L
4/XII	3	F	3.2	0.05	-	-	23m	-	-	32m	-	-	-	-	-	L
4/XII	4	F	3.7	0.1	-	-	-	-	-	10~45m	-	-	-	-	2	L
5/XII	5	M	2.9	0.1	-	-	20m	-	-	20~55m	-	-	-	-	1	L
5/XII	4	F	3.5	0.1	-	-	18m	-	-	15~60m	-	-	-	-	1	L
6/XII	2	F	2.2	0.2	-	-	-	-	-	60m	30m	-	-	-	-	L
7/XII	2	F	2.2	0.2	-	-	34m	-	-	40m	40m	-	-	-	-	L
7/XII	3	F	3.3	0.3	-	-	-	-	30m	30m	9~90m	-	-	-	2	L
8/XII	2	M	2.2	0.3	-	-	-	-	40m	25m	15~90m	-	-	-	-	L
8/XII	5	M	2.3	0.3	-	-	35m	-	80m	49m	30m	-	-	-	-	L
10/XII	5	M	2.8	0.5	-	-	-	60~90m	90m	18m	35m	60m	80m	-	1	L
11/XII	4	M	3.4	0.5	90m weak	-	24m	80m	80m	45m	40m	58m	-	-	1	L
12/XII	1	M	1.45	1.0	48m	20~108m	30~126m	34m	51m	51m	20m	62m	31m	-	3	D 6h22m
13/XII	2	F	1.75	1.0	45m	20~360m	-	30~70m	-	23m	-	80m	-	16m	3	D 20h
14/XII	3	F	3.2	1.0	40m	18~360m	24m	38m	40m	20m	20m	70m	51m	50m	2	D 16h

Note: ** AAL: Angeictasis in Auricles and Lips
L: Living, D: Death

Sinomenin hydrochloride

A Japanese researcher, Ishiwari, proved that Sinomenin hydrochloride is effective to cause peripheral perception paralysis. As reported by Mori and Hayashi, it had less paralysing effect on central algesthesia. Nevertheless, employed as infiltrative narcotic, it has strong paralysing effect on local algesthesia.

Administration by subcutaneous injection: As indicated in Table 9 and 10, after sinomenin hydrochloride was given at 0.1 ~ 0.2 ml per kilogram body weight, slight hypoalgesia occurred in 10 minutes. The most significant reactions were that red skin around palpebral fissure, auricles and apax nasi, papule urticans, extreme itching, frequent scratching, abnormality of limbs as trembling ceaselessly, and meanwhile being in excitatory state. Conclusively, sinomenin hydrochloride had no absorptive analgesia effect.

After sinomenin hydrochloride was given at 0.5ml/kg, observed were platycoria, auricles and apax nasi turning red, angeictasis, presence of papule, hypersalivation and vomiting, and slight hypoalgesia in 10 to 15 min, paralysis happening in 25 min. as no reaction when incision was made at the injection area. Nor were pain response indicated for 2.5 ~ 3 hours. Subsequently displayed were respiratory disturbance and cyanosis at apax nasi but absorptive analgesia effect was not present. After 4 hours all symptoms disappeared. Vomiting was not drastic. Salivary secretion increased, along with 1 ~ 2 times diarrhea, and urinating.

After sinomenin hydrochloride was given at 1.0ml/kg, slight dyscinesia occurred between about 37 and 93 min, without hypoalgesia, however.

Veratrin

Administration by subcutaneous injection: As shown by the 19 cases of Table 10, no reaction was observed after Veratrin was given at 0.02 mg/kg.

After Veratrin was given at 0.05 ~ 0.1 mg/kg, observed were vomiting and meanwhile being in excitatory state but absorptive algesthesia effect of Veratrin.

After Veratrin was given at 0.3 mg/kg, observed symptoms were excitement, platycoria and so.

After Veratrin was given at 0.5 mg/kg, observed were above symptoms, along with slight dyscinesia and dyspneic respiration. Absorptive hypoalgesia appeared gradually and slowly in 90 min.

After Veratrin was given at 1.0 mg/kg, extreme hypersalivation took place between 20 min. to 100 min., while drastic vomiting happened in 30 minutes as 11 times within 20 min. Dyspneic respiration was indicated. No reaction was caused when incision was made in the skin in 50 min. Nor were pain response shown but the presence of complete paralysis of algesthesia, platycoria, hyperreflexia, tremor. Motor paralysis occurred in 60 min. with only the head hold up. The subjects deceased between 6 to 20 hours after. During this period observed were 1 to 2 times diarrhea and screaming like head was tightened, dyspneic respiration and at the meantime, absorptive analgesia.

Morphine

Administration by subcutaneous injection: As indicated by the 14 cases of Table 11, after morphine was given at 1 mg per kilogram body weight, the subjects displayed uneasiness in 1.5 min., excitement, platycoria, and absorptive hypoalgesia in about 30 min., complete paralysis in 50 min. Nevertheless, they recovered to normal in 3 hours.

After morphine was given at 2 ~ 5 mg/kg by subcutaneous injection, observed were vomiting, platycoria, hypoalgesia between 10 to 30 min., and shortly paralysis. Besides, uneasiness and reflex irritation were also indicated.

After morphine was given at 10 ~ 20 mg/kg by subcutaneous injection, observed were vomiting, platycoria, and excitatory state in 30 min., absorptive hypoalgesia. The subjects recovered slightly in 5 ~ 8 hours and completely in 7 ~ 10 hours, with excitatory state remained, notwithstanding.

Fugutoxin's analgesia effect took place after dyscinesia and only did so when lethal dose was given.

The general effect of Fugutoxin is nearly the same as that of Tetrodotoxin in substance. The poisoning symptoms caused by them are fundamentally in the same order, or to say, analgesia would occur after dyscinesia rather than during the latter. As for Fugutoxin, it has stronger analgesia effect.

Table 11. Administration of Morphine hydrochloride (1%) to cat by subcutaneous injection

Time	No.	Sex	Body weight (kg)	Dose ml/kg	Absorp-tive Analge-sia	AAA*	Vomi t-ing	Platy-coria	Reflex irrita-tion	SMI*
10/VI	1	M	3.2	0.1	24-42m	190m	4m	15m	20m	-
11/VI	2	F	2.3	0.1	28-47m	210m	-		80m	-
12/VI	3	F	2.7	0.1	20-45m	120m	7m	24m	50m	-
13/VI	3	F	2.7	0.2	35-45m	120m	3-5m	11-300m	48m	-
14/VI	1	M	3.2	0.2	10m	190m	-	20m	10m	-
15/VI	2	M	2.3	0.2	28m	110m	4m	14m	50m	-
16/VI	3	F	2.7	0.5	26m	140m	2-3m	18m	32m	75m
17/VI	1	M	3.2	0.5	23-45m	200m	3-5m	22m	23m	-
18/VI	2	F	2.2	0.5	50m	190m	3m	15m	20m	84m
5/VI	2	F	2.0	1.0	53-90m	24h	3-4m	13m	27m	-
6/VI	1	M	3.0	1.0	20-40m	5h	3m	10m	20-300m	40m
7/VI	4	F	2.9	1.0	47m	6h	4m	25m	25-500m	72m
20/VI	4	F	2.8	2.0	26m	8h	2m	20m	20m	48-130m
21/VI	1	M	3.1	2.0	20m	9h30m	3-4m	25m	23m	80m

Note: * SMI: Spontaneous motor inhibition AAA: Absorptive Algesia Appearing
L: Living, D: Death m: minute h: hour

Sinomenin has no analgesia effect at all. Notwithstanding, complete analgesia appeared in the local injection area of skin. To inject Sinomenin at 0.5 ml per kilogram body weight is able to cause local complete analgesia. Therefore, it has a strong effect as an narcotic for paralyzing local perception. According to recent report by Mori and Hayashi, sinomenin also has local analgesia effect on human being.

After it was injected sinomenin, a cat indicated papul urticans in its face and extreme itching, along with other abnormal sensations. According to the report by Ijiri and Ono, one human being would also indicate anaphylactoid symptoms after he was injected sinomenin.

After Veratrin was injected at 0.5 mg per kg body weight, analgesia was observed when the subjects indicated dyscinesia. But the twice of this, 1 mg/kg, was lethal dose.

After morphine was injected at 1 mg/kg per kg body weight, the subjects indicated absorptive hypoalgesia in about 20 min., and paralysis in 40 min. This dose could cause slight vomiting 1 to 2 times and platycoria. Depending on the amount given, analgesia might took place earlier. No pain response was observed when an incision was made in the skin. The more the amount of morphine was given, the longer the period between the presence of analgesia and the reappearance of algesia.

In general, it was a sensitive method making incisions in the pain part of a cat to examine algesia. This method proved that morphine has significant analgesia effect, Veratrin has slight analgesia effect, and sinomenin has significant local paralysis effect. Nevertheless, under the same experimental conditions, with this method employed to examine the effect of Fugutoxin and Tetrodotoxin, it couldn't be proven that they have absorptive analgesia effect or local paralysis effect.

c. Clinical observation of Tetrodotoxin

Clinical observation was conducted for the effect of Fugutoxin which was given by injection to the patients who suffered neuralgia and/or other pains and seek relative treatment.

Case 1.

Kawama Mori 67 years of age Myalgia

9/II For 4 years the patient had had tension and frequent megalgia in his posterior parts of both thighs so that he was not able to fall asleep at night. His symptoms were tightness of quadriceps thigh, tenderness in intorter and walking function part of sciatic nerve, urinous albumen being moderate positive, sugar negative, BP 175 ~ 100 mm Hg.

As soon as 0.5 ml Tetrodotoxin was injected to the pressure pain point in his quadriceps of thigh, acupuncture examination was performed and observed were no hypoalgesia in skin but that muscular tension in the affected part was assuaged slightly. The patient had a sound sleep that night. He did not get up for emiction, though he used to every night before.

11/II 0.8 ml Tetrodotoxin was injected to the patient and in 30 min. he felt in his finger tips abnormality which disappeared in 6 hours. He had a sound sleep without emiction at night. In the next morning muscular tension was assuaged. BP was 160 ~ 80 mm Hg before the injection, and 147 ~ 80 mm Hg one hour after.

12/II By the same way 1.3 ml Tetrodotoxin was injected to his intorter and in 17 min. he felt abnormality in his finger tips. Meanwhile analgesia was indicated in his intorter being pressed. While walking, he felt tension in the thighs disappeared. Notwithstanding abnormalities felt at his finger tips and lips, no hypoalgesia. In 20 min. he felt thalposis in the body. BP was 155 ~ 72 mm Hg before the injection, and 147 ~ 80 mm Hg after. He had a sound sleep that night.

13/II ~ 17 ~ 18 ~ 19 ~ 20 ~ 21/II 1.0 ml Tetrodotoxin was injected to the patient each day. After the injection, in 15 ~ 20 min. he felt in the finger tips and lips abnormalities which disappeared in about 60 min. Also indicated were general thalposis, gradually disappearing

muscular tension and pain, but hypoalgesia or analgesia at any time. BP was 170 ~ 70 mm Hg before the injection, and 160 ~ 92 mm Hg after the treatment.
In general, the injected Tetrodotoxin had some therapeutic efficacy on the patient's myalgia and muscular tension, but hypoalgia was never indicated. Every time after injection, abnormalities in the lips and finger tips were displayed.

Case 2.
Tani X X 59 years of age. Myalgia
10/II The patient had an traffic accident while driving a bicycle 8 years ago. He had pain in the left upper arm so that it could not be lifted upward or turn backward. Tension was indicated in the scapular region, especially in mitroid muscle, with tenderness. Also felt were tenderness and tension in the intorter of his left thigh. His emiction was not abnormal, BP 150 ~ 82 mmHg. After 1.0 ml Tetrodotoxin was given to the quadriceps, he felt abnormality in the tongue tip and around the lips in 12 min. and 22 min. respectively. The abnormalities in the tongue tip and around lips disappeared in 30 min. and 4 hours respectively. He felt thalposis so that he had a sound sleep at night.
11/II After 1.0 ml Tetrodotoxin was given to the intorter, he felt abnormality in the tongue tip and around the lips in 7 min. and 22 min. respectively. In 2 hours these abnormalities disappeared with no other symptoms indicated. While walking, his legs moved agilely. He felt thalposis at night and had no emiction every night with sound sleep. BP was 145 ~ 72 mmHg before the injection, and 140 ~ 80 mmHg after.
12/II After 1.3 ml Tetrodotoxin was given to the intorter, he felt abnormality in the tongue tip and around the lips in 10 min. and 28 min. respectively. In 40 min. he felt itching in the upper arm, yawning continuously. These symptoms disappeared in one hour. Though it snowed heavily that day, he felt general thalposis. His legs moved agilely. He didn't need hot water bag at any night. BP was 155 ~ 95 mmHg before the injection, and 178 ~ 95 mmHg after.
13/II ~ 17/II After 1.0 ml Tetrodotoxin was given, he indicated the same symptoms as before. The treatment described above had tenderness and tension of his quadriceps and intorter removed, and nonetheless BP increased by 20 ~ 30 mmHg after the injection.
18/II Herein after 10.ml Tetrodotoxin was injected to the mitroid muscle every day. The different symptoms from before were continuous yawning and general itching. BP increased by 20 mmHg after the injection. Later the injection dose was increased to 1.5 ml. Subsequently indicated were such symptoms as yawning, nausea and dyspneic respiration, which disappeared in 2 hours. However, no absorptive hypoalgesia or paralysis happened but hyperalgia. The treatment described above was able to have pain and tension in the scapular region, and pain in the thigh removed so that the patient could walk for a long time. Its therapeutic efficacy was significant. No paralysis of algesia but abnormal sensations, yawning, nausea, itching, thalposis were observed.

Case 3.
Kawa X X 71 years of age. Sciatia, Lumbago
17/II His lumbago and pain in the thigh was caused by carrying weight 15 years ago. The pain became intense especially at back pain position(?) and caused such spasm-like megalgia that he could not fall asleep. There was pressure pain point in the walking function part of sciatic nerve. After 0.5 ml Tetrodotoxin was given respectively to the right side of lumbar region and quadriceps muscle, no reactions were indicated. BP was 145 ~ 80 mmHg before the injection, and 147 ~ 65 mmHg after.

20/II and 21/II After 1.3 ml Tetrodotoxin was injected, in 30 min. he felt abnormal in the lips and his respiration rate increased to 30/min. BP was 145 ~ 58 mmHg before the injection, and 152 ~ 60 mmHg after.
22/II By the same method 1.5 ml Tetrodotoxin was injection, he felt abnormal in the lips in 1.5 min. With acupuncture examination conducted, no hypoalgesia or paralysis was observed.
23 ~ 24 ~ 25/II After 1.3 ml Tetrodotoxin was injected continually, his algesia was not assuaged. The treatment was terminated due to inefficacy.

Case 4.
Ishikame Ro 65 years of age Mysalgia in the scapular region
26/II Megalgesia had taken place in his upper arm, especially in the scapular region for half a year. His wrist could not move up or down, nor rotate. He was not able to tie a belt or to put on a shirt by himself. At night or when having cold exposure, his pain became more drastic.
After being injected 0.7 ml Tetrodotoxin, he felt abnormal in the lips and thalposis in 2 min., shortly nausea, and vomited twice in 30 min. He also indicated dysthymia, vertigo, and abnormal sensation in the finger tips. All symptoms disappeared in 7 hours. Due to significant side effects, next injection was cancelled.

Case 5.
Test on the operator himself Tension in the scapular region
24/II After being injected 0.5 ml Tetrodotoxin, he felt abnormal in the lips and finger tips in 6 min., thalposis in 35 min., and began yawning in 40 min. All symptoms disappeared in 50 min but thalposis. He felt relieved somehow in the scapular region, with no hypoalgesia in the part of abnormal sensation. He did not feel thalposis at night.
2/VI After 0.6 ml Tetrodotoxin was given to his upper arm by subcutaneous injection, he felt the thegmesthesia of his lips and tongue tip dull and numb, but not algesia. This symptom disappeared in 90 min. while the thegmesthesia of the injected area also felt dull and numb slightly. No change happened to algesia but the tension of the scapular region was assuaged insignificantly.

Case 6.
Kusu Fuko 68 years of age Lumbago

2/VI After 0.5 ml Tetrodotoxin was given to his right upper arm by subcutaneous injection, he felt the thegmesthesia of his tongue tip dull and numb, but not the algesia. The thegmesthesia of the injected area also felt dull and numb slightly but no change happened to the algesia in the same area. The dull numbness of the tongue's perception disappeared in 90 min. Tetrodotoxin had no therapeutic efficacy on his lumbago.

Case 7.
Kame 34 years of age Tenosynovitis
17/II He had had pain in his feet and articulations of foot for two years. The pain was more intense at the time he got up and at 2:00 o'clock in the afternoon. Sometimes he was not able to walk at all. Around Achilles tendon indicated tenderness which was tough without tumefaction. After 0.5 ml Tetrodotoxin was injected into the outer side of his Achilles tendon, he felt abnormal in the foot in 10 min. and his pain assuaged insignificantly, and thalposis in 20 min. Pain disappeared, unusual feeling on lips and tongue, no analgesia indicated when algesia test was done. His pain was lessened when his feet touched the ground the next morning.

18/II ~ 19 ~ 20/II After 1.0 Tetrodotoxin was injected continually, he felt abnormal and thalposis, and insignificantly recovering in the next morning. Nevertheless, he felt pain again in the next afternoon. Therefore the treatment was terminated and the patient was transferred to department of surgery.

Case 8.

Taka x x 46 years of age Mysalgesia

5/III His pain in the left scapular region and his mysalgesia in the left greater pectoral muscle began 3 months ago, having no relevance to respiration.

After 0.5 ml Tetrodotoxin was injected into the deep part of the pressure pain point of his mitroid muscle, he felt general thalposis in 10 min.

6/III By the same method 1.0 ml Tetrodotoxin was injected. He felt abnormal around the lips and general thalposis in 10 min., abnormal in the finger tips in 20 min. with no change to the algesia. In 1 hour he indicated nausea which disappeared shortly. He had a sound sleep at night.

7/III ~ 8 ~ 9 ~ 10/III After being injected 1.3 ml Tetrodotoxin, he indicated yawning, which disappeared in 30 min., besides the above symptoms. In this case, Tetrodotoxin had therapeutic efficacy on mysalgesia with slight hedonia.

Case 9.

Tsutsumi x 64 years of age Pain in the scapular region Althralgia

10/VI His right scapular region began to ache 8 years ago, becoming so intense especially with cold exposure that it was difficult for him to get up. Besides, his lower limbs began feeling so cold three years ago that he was not able to fall asleep. Tonic tenderness was observed in his scapular region, with minim albumen and sugar negative in his emiction.

After 0.8 ml Tetrodotoxin was injected into the pressure paint point in the outer side of his right mitroid muscle, in 1.5 min. he felt general thalposis lasting till that night, while cold sensation of foot didn't disappear.

11/VI After being injected 1.0 ml Tetrodotoxin, he felt abnormal in the lips in 13 min. and in the finger tips in 23 min., and thalposis, meanwhile he yawned continuously, with thalposis in the feet. Also he felt abnormal in the tongue tip in 30 min., and nausea in 90 min. He vomited, and indicated dysthymia at that night.

12/VI ~ 13/VI After being injected 0.8 ml Tetrodotoxin, he felt insignificantly relieved in the scapular region. But the cold sensation of foot remained the same as before the injection. Due to side effects, the treatment was terminated.

As indicated by clinical experimental observation conducted in these 9 cases, in 8 of them the patients who suffered muscle tension accompanying pain, neuralgia, tenosynovitis were fundamentally cured or assuaged after the local injection of Tetrodotoxin had been given. Most patients felt abnormal in their lips, tongue tips and finger tips, and slightly dull and numb thegmesthesia, but normal algesia was observed by acupuncture examination. No analgesia effect of Tetrodotoxin on lumbago was found after it was given to upper arm by subcutaneous injection. This meant Tetrodotoxin had no remote action. The thegmesthesia at the injected local area became dull and numb while the algesia didn't but become hyperalgia. No effect on skin's algesia was observed, completely concordant with the results of the test of cats. Tetrodotoxin's analgesia effect was only indicated in local area. When Tetrodotoxin was given to local area by subcutaneous injection, the skin's algesia did not become dull and numb at all. Tetrodotoxin's remote action was so limited as only to cause dull and numb themesthesia in the lips, tongue tip and finger tips. Themesthesia became dull and numb first, similar to the results of the test of cats. The paralyzing effect of Tetrodotoxin on motor nerve is greater than that on

perceptive nerve. Administered by local injection to human beings, Tetrodotoxin is capable of assuaging muscle tension. Therefore, Tetrodotoxin has significant effect on pain.
One of human being's reactions to Fugutoxin is thalposis of body. It was in rigid February that the treatments were conducted while all patients felt warm at night. This possibly lied in peripheral angeictasis caused by injected Fugutoxin. As reported by Morijima, the analgesia effect of Sinomenin hydrochloride is relevant to its function to cause peripheral angeictasis. It is the same with Fugutoxin.
As a characteristic of intoxication by Fugutoxin, the sufferers vomit extremely frequently, reported explicitly by Fukuda and Tani. Fugutoxin usually causes nausea and vomiting when injected.
Nevertheless, a noticeable question about Tetrodotoxin is that substantial amount of carbolic anticeptic (0.5%) was added into Tetrodotoxin injection. As demonstrated by the test conducted by Kurose with samples of toad's gastrocnemius muscle, the excitation effect of carbolic acid is greater than the paralyzing effect of Tetrodotoxin. It is well known that carbolic acid has significant local analgesia effect. Therefore, carbolic acid possibly contributed to the effect of Tetrodotoxin injection during the treatments.

3. Effect on respiratory and circulatory systems

Osawa, Takahashi, Ihoko, etceteras proved with experiments that Fugutoxin is able to effect respiratory paralysis in animals, as many researchers consent to. As far as human being is concerned, respiratory inhibition and cyanosis are the primary symptoms of intoxication. According to the report by Fukuda and Tani, the direct cause for most deaths was respiratory paralysis. With marmot, dog and cat as experimental subjects, the observed general reactions to Fugutoxin were decrease in respiration rates, then hypopnea and cyanosis. Takahashi and Ihoko thought the cause to be the paralysis of medullary respiratory centre caused by Fugutoxin. However, Hayashi and Takefuji thought that respiration arrests resulted from that Fugutoxin effected the paralysis of phrenic nerve ending and other motor nerve ending. Iwakawa and Kimura carried out elaborate study on this issue. Rabbits indicated paralysis of aspneustic center after they were given Fugutoxin by subcutaneous injection; they would easily indicate paralysis of phrenic nerve ending after being given Fugutoxin by intra-venous injection. Therefore, the region to be paralyzed will be different when Fugutoxin takes effect at different speeds as by various administration routes.
Takahashi and Inoko reported that Fugutoxin's effect on circulatory system is to paralyze the center of medullary blood vessel. Kunimasa thought Fugutoxin has angeictasis effect on blood vessels. Recently Yano proved Fugutoxin does possess such effect.
Takahashi and Ihoko thought Fugutoxin has no direct effect on heart, while Ishihara believed that at the beginning it has stimulating action on heart, occurring after Flimmer and Flatter, stimulating and paralyzing the conduction system between cardiac atriums and ventricles, creating block and decreasing heart rate. Yano thought that even minim amount of Fugutoxin was able to cause the indication of heart separation. The cause of human being's death by poisoning of Fugutoxin sometimes but hardly lies in paralysis of heart.
Based on the above references, it is no doubt that Fugutoxin has such effect on both center and ending as to cause disturbance to respiration, heart and blood vessel. However, it is not recognized which of them is the leading factor. Hence, it was considered to examine directly its effect on the center around bulb and interbrain by administering Fugutoxin inside cerebellomedullary cistern and lateral ventricle. It was described in a reference that Ishihara selected such injection method administering Fugutoxin inside spinal subarachnoid space, brain matter and medullary matter. On the other hand, no studies had been known on Fugutoxin's effect on the heart of any homothermal animal. Cardiotamfour method was utilized in research

of this issue. It is known to all that carotid sinus reflex plays an important role in the pharmacological reaction of respiratory and circulatory systems. Therefore, intorcarotid injection of Fugutoxin was given before and after the removal of sinus nerve in order to examine its effect on carotid sinus reflex.

a. Test in Rabbits

To intubate a T tracheal catheter into the trachea of a subject with one end jointed with a Mareg tympania tracing respiration on black paper. To observe BP at the carotid of one side with a mercury manometer. To observe cardiomotility by Turanuku's Cardiotamfour method. Administration of Fugutoxin by iv. to rabbits without being narcotized:
After 0.0005 ml Fugutoxin per kg body weight was given by intra-venous injection to rabbits without being narcotized, no changes happened to respiration or cardiomotility.

After 0.005 ml/kg Fugutoxin was injected, blood pressure fell at the beginning and then rose. Some rabbits indicated instant elevation of BP after the injection by 30 ~ 40 mm Hg, meanwhile heart rate also increased slightly. Some rabbits indicated significant increase in respiration rate while some others decrease. It was noticeable that the elevation of BP and the polypnea and respiratory distress indicated paroxysmal during the elevation of BP. (Please refer to Fig. 1)

After 0.01 ml/kg Fugutoxin was injected, BP fell slightly and most rabbits recovered in 7 ~ 8 min. Their respiration rates decreased gradually.

After 0.5 ml/kg Fugutoxin was injection, BP fell dramatically with respiration rate decreasing. Respiration arrests occurred in 2 min. and BP continued to fall till zero in 3 min.

Administration of Fugutoxin to narcotized rabbit by iv.: Urethane was employed as narcotic. 5ml 20% urethane per kg body weight was given by subcutaneous injection. Cardiomotility was observed.

After 0.0005 ml Fugutoxin per kg body weight was given by iv., BP rose slowly and slightly. The amplitude of apex beat (peak and bottom) decreased slightly, while no change happened to that of side beat. The amplitude of heart beat also decreased with heart rate increased slightly. Respiration rate and amplitude both increased. Heart rate recovered to that before the injection in 50 min. BP rose by 10 mm Hg., the amplitude of respiration expanded, respiration rate increased. These reactions last a long time.

After 0.005 ml/kg Fugutoxin was given, BP fell by 14 mm Hg transiently, then recovered in 2 min., and rose in 3 min., rose by 6 mm Hg in 5 min. Cardiomotility: The amplitude of apex beat, especially at peak, decreased. That of side beat also decreased at both peak and bottom, but only at the time BP fell. Heart rate increased as soon as BP recovered. The amplitude of respiration reduced temporarily after the injection, and recovered shortly. (Please refer to Fig 2.)

Fig 1. A rabbit without being narcotized. 2.3 kg, male.
↑ 0.005 ml Fugutoxin injected per kg body weight.
1. Respiration 2. BP of carotid Time scale: 6s

Fig 2 A narcotized rabbit. 1.9 kg, male.
↑ 0.005 ml Fugutoxin injected per kg body weight.
1. Respiration 2. BP of carotid 3. Apex beat 4. Side beat Time scale: 6s

After 0.01 ml Fugutoxin was injected, BP fell drastically by 20 mm Hg and recovered completely in 5 min. The amplitude of apex beat reduced, especially at peak. While that of side beat varied the same as BP did, and recovered in 8 min. Heart beat increased and continued to do so after the recovery of cardiomotility. After injection the amplitude of respiration movement expanded temporarily and reduced shortly, and increased again in 8 min. At the beginning respiration rate decreased and remained reduced significantly after the recovery of BP and cardiomotility.
After 0.05 ml Fugutoxin was injected, BP fell straightforwardly by 25 mm Hg and respiration arrest occurred in 7 min. Shortly BP reduced to zero. Cardiomotility: The amplitude of apex and side beat reduced at both peak and bottom. At the beginning heart rate reduced. After the respiration arrest, it continued to decline into weak pulsation till heart arrest. The amplitude of respiration expanded, and respiration rate increased at the beginning then reduced significantly shortly. Respiration arrest took place in 7 min.
Injection of Fugutoxin into cerebello-medullary cistern: Rabbit narcotized with urethane was employed in the test.
After 0.0001 ~ 0.000125 ml Fugutoxin per kg body weight was injected into cerebello-medullary cistern, BP declined slowly and slightly while heart rate and respiration rate reduced. Some rabbits had respiration arrests about 20 min. after each was injected 0.000125 ml/kg Fugutoxin. The rabbits injected 0.0001 ml/kg Fugutoxin deceased 26 min. after they were injected another 0.0002 ml/kg Fugutoxin each.
After 0.0005 ml/kg Fugutoxin was injected, BP fell straightforwardly. The amplitude of apex beat and that of side beat both declined, while respiration amplitude and rate decreased. Respiration arrests occurred in more than 20 min. and BP fell to zero shortly. 2 rabbits deceased after being injected this dose of Fugutoxin each.
Injection of Fugutoxin into lateral ventricle: Rabbits without being narcotized were used in the test, during which BP and respiration were observed with Turanuku's Cardiometer employed for measuring cardiac volume.
No reactions were indicated after 0.0001 ml Fugutoxin per kg body weight was injected.

After 0.005 ml/kg Fugutoxin was injected, BP remained unchanged at the beginning and then increased by 10 mm Hg in 3 min. Respiration rate increased and respiration amplitude expanded in 3 min. Cardiomotility: Only heart rate reduced. Respiration rate increased to 2 times that before injection. Later heart rate declined gradually. BP fell in 1.5 min. Respiration rate decreased and so did respiration amplitude, notwithstanding, heart rate increased. BP fell significantly in 30 min. Respiration arrests occurred and BP reduced to zero in 35 min.

After 0.0005 ml/kg Fugutoxin was injected, BP remained unchanged, only respiration rate and heart rate reduced. No significant difference from that before injection was indicated. Subsequently, after 0.001 ml/kg Fugutoxin was injected, the amplitude of respiration expanded temporarily and respiration rate increased, while BP declined temporarily. In 4 ~ 5 min. BP rose by 20 mm Hg meanwhile the amplitude of respiration declined, then respiration rate reduced drastically with respiration amplitude expanding. At the beginning cardiac volume decreased, then increased along with the elevation of BP. Respiration arrests occurred in 12 min.
After 0.005 ml/kg Fugutoxin was injected, BP gradually rose by 30 mm Hg and then declined to zero in 10 min. At the beginning cardiac volume increased at both systole and diastole. Heart rate reduced. Heart arrests occurred in 20 min. At the time of injection respiration indicated temporary excitation and then its amplitude declined. The amplitude expanded in 2 min., while respiration rate increased significantly. Respiration rate reduced in 6 ~ 7 min., and then respiration arrests happened.

The effects of excitant and narcotic: After 0.005 ml/kg Fugutoxin was injected into lateral ventricle, strong respiratory inhibition and fall of BP were caused. Then 0.5 ml coramin or cornidin per kg body weight was injected. Subsequently, the inhibited respiration was instantly excited, the fallen BP rose dramatically till to normal level, and cardiomotility turned active as well. Notwithstanding, the excitation vanished in several minutes while respiratory paralysis happened, BP fell and the subject deceased.

On the contrary, Veronalnatrium can intense intoxication of a subject as to reduce its BP to zero and cause its respiration arrest.

Tetrodotoxin's effect: After 0.002 ~ 0.01 ml Tetrodotoxin per kg body weight was injected to a rabbit without being narcotized by intra-venous injection, no change happened to BP while the respiration rate increased insignificantly.

After 0.02 ml/kg Tetrodotoxin was injected, BP kept falling slightly for 2 ~ 3 min. Heart beat increased and the amplitude of respiration reduced temporarily and slightly. But some subjected did not indicate any change in their amplitude of respiration.

After 0.05 ml/kg Tetrodotoxin was injected, BP reduced significantly by 60 mm Hg and then recovered completely in 1 hour. At the beginning the amplitude of respiration expanded, then reduced in 2 ~ 3 min. Respiration rate decreased. One hour after, only respiration rate increased. Some subjects only indicated the fall of BP by 20 mm Hg, which last about 10 min.

After 0.0002 ml/kg Tetrodotoxin was injected into cerebello-medullary cistern, BP fell and the amplitude of both apex beat and side beat reduced. The amplitude of respiration reduced as well.

After 0.0005 ml/kg Tetrodotoxin was injected, BP fell drastically, respiration arrests occurred and shortly so did heart arrests.

After 0.001 ml/kg Tetrodotoxin was injected, BP rose and respiration was excited. Some subjects did not indicate any change in cardiomotility.
Only after 0.005 ml/kg Tetrodotoxin was injected by iv., BP fell transiently at the beginning and then rose. At the beginning heart rate increased, the amplitude of respiration expanded, respiration rate increased. As for some subjects, the elevation of BP and hyperpnea were indicated proxysmal.

After 0.01 ml/kg Tetrodotoxin was injected, BP would certainly fall and recover to normal in several minutes without tendency to rise. Most subjects indicated decrease in respiration rate.

It has no significant influence on Fugutoxin's effect whether to induce narcosis with urethane or not. After 0.005 ~ 0.01 ml/kg Fugutoxin was injected to a narcotized rabbit, the fall of BP was insignificantly intensified and the elevation insignificantly weakened. After 0.005 ml/kg Fugutoxin was injected, BP and respiration would not indicate proxysmal excitation.

After 0.05 ml/kg Fugutoxin was injected, BP fell quickly and cardiomotility was inhibited. Respiration rate reduced significantly and respiration arrest occurred shortly with BP falling to zero. This dose is confirmed lethal dose.

After 0.0001 ml/kg Fugutoxin was injected into cerebello-medullary cistern, no reaction was indicated. After 0.0005 ml/kg Fugutoxin was injected, BP rose slightly while respiration rate and heart rate reduced. Some subjects indicated respiratory excitation at the beginning but then deceased in 35 min. After 0.001 ~ 0.005 ml/kg Fugutoxin was injected, BP rose and respiration was excited, but then BP fell drastically and respiration was inhibited till respiration arrest occurred in 10 min.

As demonstrated by the above results from the test of rabbit, after small amount of Fugutoxin was injected, respiratory excitation, increase in heart beat, and elevation of BP were indicated at the beginning. By comparison with that of administration by iv., Fugutoxin's effect by direct injection into brain center was greatly intensified. Or to say, the ratio of the least effective doses of these two methods is 1: 50, and the ratio of their lethal doses is 1: 100. Conclusively, Fugutoxin's action point is not on the endings but on the central nerves.

It is noticeable that Fugutoxin has different effects between the administration by injection into cerebello-medullary cistern and that by injection into lateral ventricle. Generally, Fugutoxin has stronger effect by injection into cerebello-medullary cistern than by injection into lateral ventricle. It is to say, reactions will be indicated when 0.0001 ml/kg Fugutoxin is injected into cerebello-medullary cistern, and some subjects will decease when 0.000125 ml/kg Fugutoxin is injected. While, no reactions will be indicated when 0.0001 ml/kg Fugutoxin is injected into lateral ventricle but 0.0005 ml/kg, at which death is often caused as well. The more significant difference is that the fall of BP and respiratory inhibition are more remarkable, and elevation of BP and respiratory excitation are rarely indicated when Fugutoxin is injected into cerebello-medullary cistern. As for injection of Fugutoxin into lateral ventricle, the elevation of BP and respiratory excitation will certainly take place at early stage and then so will the fall of BP and respiratory inhibition. Based on this result, Fugutoxin primarily effects center, acting on the center near to interbrain which is close to lateral ventricle, inducing elevation of BP and respiratory excitation, as well as acting on centres of medulla oblongata which is close to cerebello-medullary cistern, inducing fall of BP and respiratory inhibition.

Fugutoxin's effect is not to induce central excitation but central paralysis. Therefore, central excitant can introduce antagonism against Fugutoxin's effect, while central narcotic can introduce synergism. Conclusively, by intra-venous injection, Fugutoxin paralyzes the blood pressure center and the respiratory inhibiting center near interbrain at the beginning so as to induce elevation of BP and respiratory excitation. Shortly blood vessels and respiratory center in medulla oblongata are directly paralyzed so as to induce fall of BP and respiratory inhibition.

b. Test in Dogs

Test procedure: To administer Fugutoxin by iv. at femoral vein or by injection into general carotid to dogs which have been narcotized by mixed anesthesia of urethane and morphine. To intubate a T tracheal catheter into the trachea of a subject with one end jointed with a Marey's Tambour tracing respiration on black paper. To observe BP with a mercury manometer for femoral artery or cerebral arteries.

Administration of Fugutoxin by iv.: After 0.01 ml Fugutoxin per kg body weight was injected into the femoral vein, BP rose transiently at once by 26 mm Hg and recovered to normal in 1 min. without significant change. Sometimes BP fell slightly. Heart rate increased during both the elevation of BP at the beginning and the fall after. During the elevation of BP the amplitude of respiration expanded temporarily and shortly recovered to what it had been. In 2 ~ 3 min. the

amplitude of respiration expanded again, and more significantly especially during the fall of BP. At the beginning respiration rate increased, and more significantly especially during the fall of BP as well. A subject generally would recover completely having been injected Fugutoxin at this dose.

After 0.03 ml/kg Fugutoxin was injected into the femoral vein, BP rose transiently and slightly, then fell transiently, and recovered to normal later. Heart rate increased transiently. So did the amplitude of respiration which recovered to normal in 1 min.

Respiration rate became less than that before the injection (Fig. 3). 1.5 min. after, 0.03 ml/kg Fugutoxin was injected to the same subject. BP didn't rise but fell slowly, and recovered to normal in 1 min. Heart rate increased, and recovered to normal in 10 min. The amplitude of respiration increased transiently, so did the respiration rate which then decreased. 25 min. after the second injection, 0.05 ml/kg Fugutoxin was injected into the femoral vein of the same subject. Subsequently, BP fell gradually by 50 mm Hg. Heart rate increased transiently and then reduced, till to zero 4 min. after. The amplitude of respiration expanded and the respiration rate decreased with the fall of BP. The amplitude of respiration reduced significantly in 2 min. and respiration arrest occurred in 4 min.

After 0.05 ml/kg Fugutoxin was injected into the femoral vein, BP rose transiently by 20 mm Hg. 30 seconds later, BP kept falling slowly. Heart rate increased instantly after the injection, and accelerated with the fall of BP. At the beginning respiration rate increased temporarily and then reduced gradually. No death was caused but fall of BP and respiratory inhibition enduring a considerably long time.

After 0.06 ml/kg Fugutoxin was injected into the femoral vein, BP rose transiently by 14 mm Hg at the beginning and then fell drastically. 1 min. after, BP continued to fall gradually, and fell to zero shortly after respiration arrest. 4 ~ 5 min. after the injection, or to say, during the drastic fall of BP, heart rate increased significantly, and reduced before death. 1 min. 30 sec. After the injection, respiration rate increased transiently, shortly the amplitude of respiration reduced, and then respiration rate decreased till respiration arrest in 20 min.

Generally speaking, being given at 0.01 ~ 0.03 ml/kg to dogs by iv., Fugutoxin had such effect as to induce transient elevation of BP at the beginning, then transient fall, later elevation again. Heart rate increased slightly with no significant change in respiration. Some subjects indicated exaggerated respiration. After 0.05 ~ 0.06 ml/kg Fugutoxin was given, BP indicated transient elevation at first, and then continuous fall. Especially, death occurred shortly after 0.06 ml/kg Fugutoxin was injected. Heart rate would mostly increase, and would reduce only when BP fell drastically and death was close. The amplitude of respiration expanded transiently, then reduced after respiration rate increased, and shortly respiration arrest occurred.

Administration of Fugutoxin by injection into general carotid: Afterwards Hering and Hegmans believed that in carotid sinus exists BP receptors and chemical receptors which can induce cardiac reflex and respiration reflex. Uramoto, Harada, Zentokoro, etceteras conducted research in this issue. The following study was carried out on the action of carotid sinus based on Zentokoro's method.

After 0.03 ml Fugutoxin per kg body weight was injected, BP rose instantly and recovered to normal in 1 min. Heart rate increased with the elevation of BP, and endured a considerably long time. After the injection, the amplitude of respiration movement expanded instantly and

respiration rate reduced temporarily, then recovered to normal immediately. 85 min later, BP remained normal while heart rate still increased slightly (Fig. 4). With the carotid sinus nerve of the same subject removed, Fugutoxin of the same amount as the previous injection was injected into the carotid 90 min. after the first injection. At the beginning, BP rose transiently, then fell immediately by 70 mm Hg. At first heart rate indicated no change, then reduced slightly at a slow pace. The fallen BP recovered slowly in several min., and remained insignificantly lower than normal in 1 hour. Heart rate reduced as well (Fig. 5). Vagotomized in 1.5 hours, the subject did not indicate elevation but fall of BP. Respiration rate reduced significantly. Inspiration phase expanded temporarily and then recovered slowly. 2 min. later, 0.03 ml/kg Tetrodotoxin was injected into the carotid, at first BP fell, heart rate reduced, the amplitude of respiration expanded at the early stage of the fall of BP, respiration rate increased, and recovered to normal shortly. 5 min. later, complete respiration arrest occurred when heart rate reduced drastically with BP close to zero, and did not halt until 9 min. later.

By comparing the result of the administration by injection into carotid with that by injection into femoral vein, it could be concluded that there was no difference between their action strengths or their action duration (with the normal presence of sinus nerve). This possibly resulted from Fugutoxin's effect on the chemical receptors of carotid sinus. By comparing the reaction to the second injection of 0.03 ml/kg Fugutoxin into carotid with carotid sinus nerve removed with that to the second injection by iv., indicated were that BP fell significantly, heart rate did not increase but decrease, and respiration rate decreased as well. Conclusively, Fugutoxin had no direct effect on chemical receptors, however, Fugutoxin effected change in BP, induced reflex of blood pressure receptors so as to procure secondary increase in heart rate and secondary polypnea and distress of respiration.

Fig. 3 A narcotized dog. 13.5 kg, male.
↑ 0.03 ml Fugutoxin per kg body weight injected into femoral vein.
1. Respiration 2. BP of carotid Time scale: 5s

Fig. 4 A narcotized dog. 10.0 kg, male.
↑ 0.03 ml Fugutoxin per kg body weight injected into general carotid.
1. Respiration 2. BP of carotid Time scale: 5s

Fig. 5 A narcotized dog with carotid sinus nerve removed, 10.0 kg, male.
↑ 0.03 ml Fugutoxin per kg body weight injected into general carotid.
1. Respiration 2. BP of femoral artery Time scale: 5s

4. Effect on gastrointestinal tract (esp. vomitory effect)

a. Vomiting

The most significant symptom of Fugutoxin's effect on gastrointestinal tracts of cat and dog is, as described in the chapter about Fugutoxin's general effect, vomiting. As Takahashi and Ihoko reported, this symptom was commonplace. It was thought by some people that vomiting was not only the initial but an infallible reaction to Fugutoxin. The investigation carried out by Fukuda and Tani proved that vomiting is the most common symptom that has occurred. Nausea and vomiting are regularly caused by Fugutoxin injected.

50% vomitory dose and vomiting time displayed as following table:

Experimental animal	By subcutaneous injection		Per os	
	Vomitory dose	Incipient time	Vomitory dose	Incipient time
Cat	0.02 ml	10m ~ 40m	0.2 ml	30m ~ 35m
Dog	0.01 ml	4m ~ 15m	0.7 ml	60m ~ 70m

As for cat or dog, the 50% vomitory dose by subcutaneous injection is much less than that per os. The ratio of these two doses is 1: 10 for cat and 1: 50 for dog. The incipient vomiting time by subcutaneous injection is 10 min., or no later than 30 min., while that per os is longer as about 69 min. Based on these indications, vomiting is not caused by stimulation to gastric mucosa but by absorbed Fugutoxin acting on vomiting center. The following test was performed in order to recognize the mechanism of action further.

The vegotomization factor or atropine's action: Vegotomized at two sides of its neck, a dog was given 0.03 ml/kg Fugutoxin by subcutaneous injection. It vomited in 10 min. The frequency of vomiting was much less than the normal condition (only Fugutoxin injected), and the vomiting endured a shorter duration as 20 ~ 40 min.

25 ~ 30 min. after 0.2 ~ 0.5 mg atropine per body weight was injected, 0.02 ~ 0.04 ml/kg Fugutoxin was injected. Subsequently, the subject vomited in 10 min. After 10 ~ 15 ~ 20 mg/kg atropine was injected, the dog was induced into excitatory state in 20 ~ 30 min., not quiet, barking from time to time. Then 0.02 ~ 0.03 ml/kg Fugutoxin was injected, it vomited in 10 min., similarly as above. However, the frequency of vomiting was much less than the normal condition, with a shorter duration. The subject fell asleep in 30 min. due to the effect of atropine.

The above results suggested that the vomiting caused by Fugutoxin will not be relieved of by vegotomization or injection of atropine, but the frequency and the duration of vomiting will be reduced.

Narcotic's action: Hexobarbital sodium: 20 mg Hexobarbital sodium per kg body weight was injected to a dog by iv. and finished within 2 ~ 2.5 min. The dog snored immediately, induced into narcosis with muscular tension declined. Then 0.03 ml/kg Fugutoxin was given by subcutaneous injection, the dog was induced into narcosis in 9 min., meanwhile it vomited. About 1 hour later, it vomited repeatedly. Under this condition, it recovered from narcosis 10 ~ 20 min. after the injection, and it was able to

lift its head in 1.5 min. It was the same with several other cases of the test. This is to say, hexobarbital sodium was not able to inhibit vomiting but to reduce the frequency and retard the incipient time of vomiting.

Morphine: Morphine is able to cause vomiting itself. The kind of vomiting had been thought to be peripheral before, but Magnus believed it to be central. Egglaston and Hatcher used dogs to perform a test in which dogs were caused to vomit even though their internal organs were all removed. Having been injected morphine by Egmond, the dogs would not vomit any more after 3 ~ 4 times vomiting. Leake believed the reason to be that Brechcentrum is stimulated and turns to be paralyzed. During the test, within 3 ~ 7 min. after the injection of morphine, 2 ~ 3 times vomiting occurred and then not any more.

After 0.5 ~ 1.0 mg morphine per kg body weight was injected and one hour after the subject halted vomiting, 0.03 ml/kg Fugutoxin was injected. Subsequently the subject vomited in another 8 min. with a much less frequency than that without morphine injected, only 1 ~ 2 times.

After 2.0 ~ 3.0 ~ 5.0 ~ 10.0 ~ 15.0 mg/kg morphine was injected, 0.03 ml/kg Fugutoxin was injected, but no vomiting occurred.

The vegotomization factor or narcotic's action: After a dog was narcotized with hexobarbital sodium, vegotomization was conducted at both sides of its neck. Then 20 mg hexobarbital sodium was injected. While the dog was in narcosis, 0.03 ml/kg Fugutoxin was given by subcutaneous injection. The dog did not indicate vomiting during the following 2 ~ 3 hours. Narcosis disappeared 15 ~ 20 min. after the injection of hexobarbital sodium. The dog awaked completely in 30 min. and was able to eat.

Conclusively, after vegotomization, the injection of hexobarbital sodium is able to inhibit the vomiting induced by Fugutoxin as a subject will not vomit even when it awake from deep narcosis.

Based on above test results, after vegotomization at both sides, Fugutoxin will still induce vomiting, which is not caused by stomach reflex. It is to say, Fugutoxin induces vomiting by acting on vomiting center so as to cause the movement of abdominal wall muscle and respiratory muscle. The injection of morphine not less than 2 mg is completely able to inhibit Fugutoxin from inducing vomiting. Hexobarbital sodium of general narcotic dose is not able to inhibit Fugutoxin from inducing vomiting of normal dogs, but to reduce the frequency and strength of vomiting. After vegotomization, Hexobarbital sodium can inhibit vomiting definitely. The vomiting can also be inhibited by the administration of atropine, which does not act on the perceptive teleneurons in internal organs but only paralyze the motor teleneurons subordinated to vagus nerve. This is proved by the heart reflex test conducted by Satotsubo. That atropine can inhibit vomiting to some extent may lie in eccentric fibrous paralysis, stomach contraction and hypotonia. That vomiting induced by Fugutoxin can also be inhibited after vegotomization may result from the block of eccentric communication branch with vagus nerve.

Notwithstanding the vomiting induced by Fugutoxin is central, it is still difficult to prove Fugutoxin's effect on vomiting center at present. It is generally recognized that Fugutoxin has paralyzing effect on vegetative centres as of blood vessel, respiration and so, but it still remains in dark whether Fugutoxin has excitatory action on vomiting center only.

Similarly, although it has center-paralyzing effect, morphine induces vomiting at the early stage. Some researchers believe that its mechanism of action is that the vomiting center inhibit paralysis, or the relief of the inhibition excites the vomiting center. As observed in some morphine analgesia test of cat, after various doses of morphine was given, in most cases the cats vomited 2 ~ 3 times in 2 ~ 5 min. after the injection and halted vomiting in 10 min. The injection doses were 2.0 ~ 5.0 ~ 10.0 ~ 20.0 mg/kg. Notwithstanding the significant differences between these doses, no difference was found between the vomiting situations. This result indicated that morphine's vomiting action is not excitatory to vomiting center but a premature symptom due to relief of inhibition.

Conclusively, the vomiting caused by Fugutoxin is the same as that by morphine, being generated by excitement resulted from the relief of the inhibition on vomiting center. Especially at the early stage of Fugutoxin's effecting, the elevation of BP and respiratory excitation are caused by the relief of inhibition. Therefore it is the same with the action on vomiting center. The difference between Tetrodotoxin and morphine is that Tetrodotoxin has no direct paralyzing effect on vomiting center. This can be proved by the fact that the frequency of vomiting induce by Fugutoxin increases with Fugutoxin's doses.

That hexobarbital sodium is able to abate vomiting indicates that the vomiting induced by Fugutoxin does not result from central excitation. However, hexobarbital sodium inhibits vomiting center while Fugutoxin inhibits paralysis of the center so that vomiting is abated.

One of Fugutoxin's effects is to generate hypersalivation. This is somehow relevant to vomiting. In Ishihara's test, Fugutoxin was given to cats by subcutaneous injection or iv., it had no effect on hidropoiesis or salivary secretion. When local injection of Fugutoxin was performed, the secretory fiber was paralyzed. In the tests with cats and dogs employed, Fugutoxin was able to increase salivary secretion. With marmot employed in the test, Fugutoxin did not induce vomiting but increase in salivary secretion with the hair around marmot's mouth sopped. Similarly to many vomitories, Fugutoxin induces vomiting and increase in salivary secretion at the same time.

b. Movement of intestine canal

Based on Yamamoto's method, the intestine canal of a narcotized rabbit was drawn out. The movement of the intestine canal was recorded by graphical method after Fugutoxin was injected to the rabbit by iv.

After 0.001 ~ 0.005 ~ 0.01 ml/kg Fugutoxin was injected, the contraction of the intestine canal abated, the relaxation did not go well, and the amplitude reduced. These reaction endured about 5 min. and then were all recovered (Fig. 6).

Fig. 6 In-vitro intestine canal of a rabbit, 2.1 kg, male
0.1 ml/kg Fugutoxin injected by iv.

After 0.05 ml/kg Fugutoxin was given, the tensity of the intestine canal declined, the amplitudes of peristalsis and pendular movement reduced. This dose was lethal dose.

Extraction test of intestine canal:
Based on Magnus's method, the intestine canal of rabbit was used in the test.

No reaction occurred upon administration of Fugutoxin at the concentration of 10^{-5}, while the contraction of the intestine canal declined at 10^{-4}, as well as the tensity reduced slightly. The contraction declined and the tensity reduced significantly at ½ 10^{-3} or 10^{-3}. The declination of the contraction disappeared in several minutes.

After Fugutoxin at 10^{-2} was given, the pendular movement halted temporarily and then slowly recovered. After Fugutoxin at 10^{-3} or 10^{-2} was given and subsequently the declination of contraction disappeared, the same amount of Fugutoxin was given again at twice the concentration previously given when the contraction did not decline. Despite that the concentration of Fugutoxin of the second administration was higher, the declination of the contraction was slighter than that during the first administration.

Effect on autonomic nerve:
Having being given Fugutoxin and subsequently indicated declination of intestine contraction, the extracted sample was given pilocarpine at 10^{-6} and acetylcholine at 10^{-7}, and then its tensity increased with indication of strong contraction excitation. Therefore, Fugutoxin has no paralyzing effect on parasympathetic teleneutron.

After atropine at 10^{-5} and then Fugutoxin at 10^{-3} were given, the intestine canal indicated declination of contraction at the beginning, same as a normal intestine canal did.

With ergotaminine at 10^{-5} injected and then the inhibition action of adrenalin disappeared, the intestine canal was given Fugutoxin at 10^{-3} when the indicated reactions were the same as that under normal conditions. Therefore, the declination of intestine canal's contraction caused by Fugutoxin did not result from excitation of sympathetic nerve.

In general, Fugutoxin has direct inhibition effect on intestine canal and its effective concentration is more than 10^{-4} for extracted intestine canal.

After Fugutoxin effecting, pilocarpine and acetylcholine indicate excitatory action.

After ergotaminine effecting, Fugutoxin usually indicates inhibition action. Atropine has no effect at all. However, Fugutoxin has no effect at all when it is given again.

Fugutoxin has inhibition effect on in-vitro intestine canal, and is effective being given at minim amount by comparison with extracted intestine canal, and the mechanism of action is similar with the latter. This may result from Fugutoxin's direct paralyzing effect on the smooth muscle of intestine canal. The test carried out by Ishihava with extracted intestine canal from rabbit proved this inhibition effect of Fugutoxin. From this perspective, Fugutoxin's effect on smooth muscle should not be neglected.

Summary

(1) The injection of lethal dose of Fugutoxin to a marmot will not cause motor paralysis. The subject will indicate convulsion before death. A dog will not indicate convulsion, but no severe motor paralysis either. As for a cat, the spontaneous movement will be paralyzed completely with muscular tensity reduced but reflex remained.

(2) It is a sensitive test method stimulating the edge of the incision on a cat's back so as to observe the local algesia paralysis induced by pantocain. Fugutoxin does not effect local analgesia. Remote analgesia will appear only when given lethal dose or approximately lethal dose of Fugutoxin causes severe intoxication, while hyperalgesia will be indicated when small dose is given.

(3) It is noticeable that the limbs of a cat will tingle and indicate abnormal sensation in the endings when minimal dose of Fugutoxin is given. The same symptoms will occur when sinomenin hydrochloride is injected to a cat. A human being will indicate dull and numb perception in the tongue tip, the lips and the finger tips when he is injected Fugutoxin.

(4) Fugutoxin by respectively local injection has analgesia effect on human being's myalgia, neuralgia and tenosynovitis. But it doesn't induce remote analgesia action when it is injected to other regions of human body. In spite of the indications of dull and numb perception in tongue tip, lips and finger tips, algesia will not turn dull and numb. On the contrary, it will turn into hyperalgia sometimes.

(5) As for marmot and cat, they will indicate hyperemia in the auricles and the lips when small doses of Fugutoxin is given by subcutaneous injection. A human being injected Fugutoxin will not feel cold in the lower limbs and the lumbar region at frigid night. This may result from angeictasis of skin caused by Fugutoxin.

(6) Minim amount of Fugutoxin given to a rabbit by iv. can induce slight respiratory excitation, increase in heart rate and elevation of BP. Narcosis by urethane can lessen this excitatory effect. A slightly larger dose of Fugutoxin injected can cause insignificant fall of BP which will be recovered. A even larger dose injected can cause respiratory inhibition, decrease in heart rate and fall of BP.

(7) Fugutoxin given by injection inside cerebello-medullary cistern or inside lateral ventricle is 50 times as effective as that by intra-venous injection. In this case the effect is mainly

central. The lethal dose by central injection is 1/100 that by iv. The death by poisoning of Fugutoxin results from central paralysis.

(8) Fugutoxin injected inside cerebello-medullary cistern will induce respiratory inhibition and fall of BP immediately. Fugutoxin injected inside lateral ventricle will cause transient respiratory excitation, increase in heart rate and elevation of BP at the early stage, and then respiratory inhibition and fall of BP. Barbital has synergistic action while aramine has inhibiting action to this inhibition effect. Fugutoxin has paralyzing action on medulla oblongata centres of respiration, blood vessel and heart, and at least transient excitatory action on interbrain center. This may result from the relief of the inhibition on these centres.

(9) The effect on respiration and BP by Fugutoxin injected into general carotid is not different in quality or quantity from that by intra-venous injection. Fugutoxin has no direct action on the chemical receptors of carotid sinus. Notwithstanding, when BP falls, the pressure receptors will be stimulated and reflexively induce respiratory excitation and increase in heart rate.

(10) Fugutoxin injected to a dog or a cat can cause vomiting, which is an incipient toxic symptom as well as a definite symptom of the dog or the cat. Hypersalivation will occur around the vomiting.

(11) Vomiting can't be prevented from occurring by vegotomization or injection of atropine. However, morphine is able to do so completely. Hexobarbital sodium is able to abate vomiting. The vomiting is neither peripheral nor reflexive but central, and most likely results from the relief of the inhibition on vomiting center.

(12) Fugutoxin given at small amount can inhibit the movement of intestine canal. Most likely it has direct inhibiting action on smooth muscle.

(13) The effective dose of Fugutoxin by iv. has no significant difference from that by subcutaneous injection. Despite that hypodermis can absorb Fugutoxin quickly, the ratio of the effective dose per os and that by subcutaneous injection is 1:10 for marmot or cat, and 1: 50 for dog. Alimentary canal absorbs Fugutoxin much more slowly than hypodermis does, especially when Fugutoxin is mixed with feed and given per os.

(14) The course of intoxication of Fugutoxin for a dog or a cat develops so fast as some subjects died in 3 ~ 4 hours and some others recovered in 3 ~ 4 hours. However, some cats didn't die from persisting poisoning until several days or more than 10 days later.

Conclusions

The course of intoxication by Fugutoxin develops fast for animals as it will reach climax of intoxication in 3 hours after the administration. Many animals recovered from intoxication. Nevertheless, some cats often died from persisting poisoning. The cause of death was paralysis of central respiration and paralysis of blood vessel nerve. At the early stage of intoxication, respiratory and circulatory organs indicated transient excitation. This may result from the relief of the inhibition on the centres of respiration, blood vessel and heart. The incipient symptom of intoxication by Fugutoxin is vomiting for a cat or a dog, and it is definite to happen. This may also result from the relief of the inhibition on vomiting center. Tests in animals proved that Tetrodotoxin does not possess remote analgesia effect, nor local analgesia effect. Neither has Tetrodotoxin remote analgesia effect on human being. Its local analgesia effect results from its paralyzing action on perceptive nerve.

Fugutoxin is able to induce peripheral motor paralysis. In this respect it is similar to curarine. In the respect of the paralyzing effect on centres, it is different in quality from curarine. And in the respect of its local paralyzing effect, it is totally different from cocaine. Its remote analgesia effect is neither similar to that of morphine. Fugutoxin has peripheral and central paralyzing effect. Its effect on respiration is characterized by central paralysis. In this respect, its effect is similar in quality to that of magnesian ion.

References (omitted)